BIFUNCTIONAL SYNZYMES VIA ALTERNATING COPOLYMERIZATION

BY

DAVID PAUL VANDERBILT

A DISSERTATION PRESENTED TO THE GRADUATE COUNCIL OF
THE UNIVERSITY OF FLORIDA
IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE
DEGREE OF DOCTOR OF PHILOSOPHY

UNIVERSITY OF FLORIDA

1982

To my parents:

John and Betty Vanderbilt

who made it all possible.

To my aunt and uncle:

John and Helene Taras

who instilled in me

an appreciation of chemistry.

ACKNOWLEDGEMENTS

I would like to thank Dr. George B. Butler for the opportunity to work under his tutelage for the past three years. His encouragement and guidance is sincerely appreciated. I also thank the members of my supervisory committee.

Special thanks are extended to Dr. Kurt G. Olson, Dr. Huey Pledger, Jr., Dr. Roy King and Dr. Thomas Baugh for invaluable advice and assistance. I also thank Ms. Patty Hickerson for the skillful typing of this manuscript. I am especially indebted to my friends outside of chemistry for keeping me sane.

Financial support for this work from the National Science Foundation (Grant DMR 80-20206) is gratefully acknowledged.

TABLE OF CONTENTS

PAGE

LIST OF TABLES

LIST OF FIGURES

Abstract of Dissertation Presented to the
Graduate Council of the University of Florida
in Partial Fulfillment of the Requirements for the
Degree of Doctor of Philosophy

BIFUNCTIONAL SYNZYMES VIA
ALTERNATING COPOLYMERIZATION

By

David Paul Vanderbilt

December 1982

Chairman: Dr. George B. Butler
Major Department: Chemistry

The synthesis, characterization and evaluation of bifunctional synthetic enzymes (synzymes) via alternating copolymerization was carried out. It was desired to obtain copolymers containing alternating placements of complementary functional groups to see if "cooperativity" between the groups (in the hydrolysis of ester substrates) could be greater than in a random copolymer containing the same groups.

To this end, the following bifunctional alternating copolymers were synthesized: N-(β-vinyloxyethyl)imidazole -- N-hydroxymaleimide (53), isoeugenol -- N-[2-(4-imidazolyl)ethyl] maleimide (59), and 2-propenylphenol -- N-[2-(4-imidazolyl)ethyl] maleimide (60). These copolymers were evaluated as catalysts in the hydrolysis of p-nitrophenyl acetate (PNPA) or 2,4-dinitrophenyl benzoate (DNPB). No cooperativity between imidazole and hydroxamic acid or imidazole and phenol groups was observed in the hydrolysis of activated esters, leaving unresolved

the premise that bifunctional alternating copolymers should make better catalysts than bifunctional random copolymers.

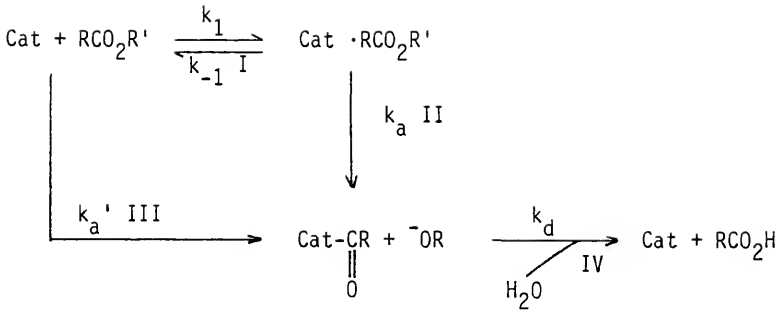
CHAPTER I

INTRODUCTION

A great deal of attention has been given to the understanding of the catalytic properties of enzymes.[1] Enzymes are globular proteins (polyamino acids) which catalyze most of the chemical reactions in living organisms. Recently, we have begun to understand the mechanisms by which enzymes catalyze organic reactions in terms of the transition state theory. As man's knowledge of enzyme mechanism has grown, so too has his wish to synthesize artificial enzymes or "synzymes." Synzymes have shown considerable utility as probes of enzyme kinetics.[2] These synthetic enzymes should emulate the desirable characteristics of natural enzymes, i.e., show high selectivity and high efficiency (rate enhancement) toward the substrate molecule. To date, both characteristics have been incorporated into synthetic polymers to some degree. As this work deals with the synthesis and evaluation of synzymes, a brief discussion of recent developments in the field follows.

Kinetic Scheme for Esterolysis

Enzymes are capable of catalyzing a great variety of chemical reactions; one of the most studied of these is the esterolysis (ester hydrolysis) reaction. In general, the kinetic scheme for the hydrolysis of an activated ester by a catalyst can be represented as follows:[2b]



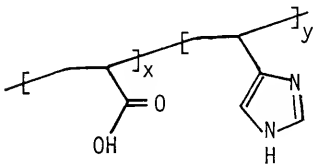
where Cat is the catalyst, and RCO_2R' is the ester substrate. Step I represents an equilibrium between free catalyst and free substrate and a catalyst-substrate complex or Michaelis complex. This step is assumed to be rapid and reversible with non-covalent binding forces holding the complex together. The actual hydrolysis steps then take place via acylation of the catalyst (II) and subsequent deacylation (IV). This pathway is believed to be important in the case where the catalyst is a natural enzyme. Alternatively, acylation of the catalyst may occur via a bimolecular reaction (III), in which no pre-association of catalyst and substrate has taken place. This pathway is followed in the case of small molecule catalysts and many synzymes. Catalysis by a synzyme might follow both reaction pathways simultaneously.

Effective Binding

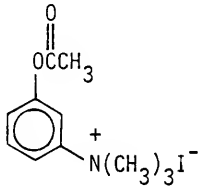
Effective binding between catalyst and substrate prior to the acylation step plays a key role in providing high catalytic activity. This pre-association step greatly increases the esterolysis rate by increasing the concentration of substrate at the active site of the

catalyst. Furthermore, acylation can take place via an intramolecular reaction (II) rather than by the much slower intermolecular pathway (III). At least four types of binding forces have been identified in the pre-association process: coulombic interactions, hydrophobic interactions, hydrogen bond formation, and charge-transfer interactions. The most important factor determining the catalyst's effectiveness, however, is that the binding take place at a site which is favorable for the subsequent acylation reaction to occur.

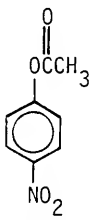
An example of coulombic interactions as the mode of polymer-substrate binding has been demonstrated by Overberger and Maki.[3] Poly[4 (5)-vinylimidazole-co-acrylic acid] (1), containing an excess of acrylic acid units (53.7 mol%) and therefore having an excess of anionic sites, hydrolyzed positively-charged 3-acetoxy-N-trimethylanilinium iodide (ANTI) faster than neutral p-nitrophenyl acetate (PNPA), which in turn was hydrolyzed faster than the negatively-charged substrate 3-nitro-4-acetoxybenzoic acid (NABA).



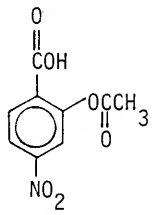
1



ANTI



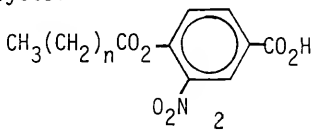
PNPA



NABA

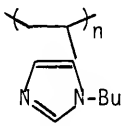
This study also demonstrates a certain degree of selectivity shown by the catalyst toward the substrate.

Favorable binding by hydrophobic interactions has been demonstrated by Klotz and Stryker.[4a] They found that partially lauroylated poly(ethylenimine) catalyzed the hydrolysis of PNPA at a faster rate than did poly(ethylenimine) itself. Indeed, the most effective synzyme studied to date is a dodecylated poly(ethylenimine) containing imidazole residues. This derivative was found to approach α-chymotrypsin in catalytic activity.[4b] On the other hand, Overberger and Smith[5] studied the effect of varying the chain length of substrate (2) using poly(1-butyl-5-vinylimidazole) (3) and poly(1-methyl-5-vinylimidazole) (4) as catalysts.

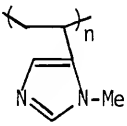


2

n = 0, 5, 10, 16



3

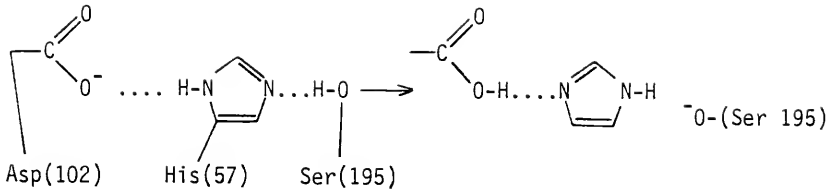


4

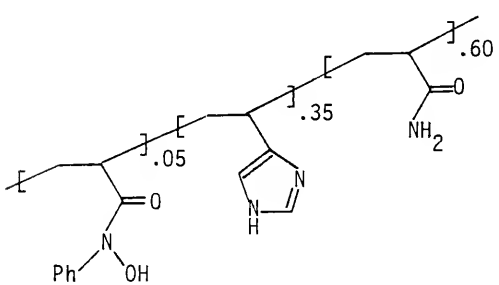
It was found for both (3) and (4) that (2, n = 16) was hydrolyzed at a faster rate than (2, n = 0).

Chemistry at the Active Site--Cooperativity

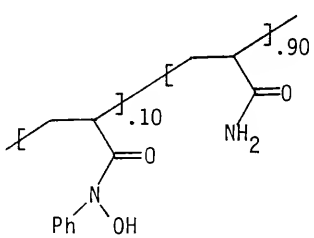
As we have seen above, in order to observe a significant rate enhancement in esterolysis reactions the substrate must first be bound to the polymer near or at the active site. Only after complexation has occurred do the actual hydrolysis steps take place. Synzyme esterolysis has been observed to proceed with or without a complexation step. Studies with α-chymotrypsin (a serine proteinase consisting of 245 amino acid residues) have shown that a serine O^- anion is responsible for catalytic acylation of substrate.[1]



The serine hydroxyl group is activated for the acylation reaction by the scheme shown above, dubbed a "charge relay system," in which the imidazole moiety plays an integral role in lowering the activation energy for catalysis. A variety of cooperative effects among the functional groups responsible for catalytic action is common in natural enzymes. The synthetic chemist has also sought to take advantage of cooperativity in order to produce more efficient synzymes. A good example of bifunctional cooperation utilizing a molecular relay system was demonstrated by Kunitake and Okahata.[6] These workers found that the rate of hydrolysis of PNPA was 1000 times faster using a terpolymer N-phenylacrylohydroxamate : 4(5)-vinylimidazole : acrylamide (5) compared with N-phenylacrylohydroxamate : acrylamide copolymer (6) as catalysts.



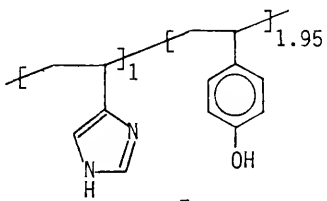
5



6

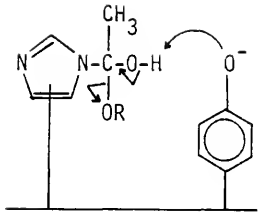
The acylation step was demonstrated to occur primarily via the hydroxamate anion, which is known to be a highly nucleophilic species. It is also known that decomposition of an acylhydroxamate is a slow process; the fact that 5 is a much better catalyst than 6 implies that imidazole is catalyzing deacylation of the acylhydroxamate intermediate either acting as a nucleophile or general base.

Another example of a bifunctional catalyst exhibiting cooperativity is a 1:1.95 copolymer of 4(5)-vinylimidazole and p-vinylphenol (7).[7a] This copolymer was 63 times as efficient as imidazole for the hydrolysis of ANTI at pH 9.1, and 10.6 times as effective as imidazole vs. PNPA at the same pH. Phenol, poly(p-vinylphenol), poly[4(5)-vinylimidazole] and a 1:0.48 copolymer of 4(5)-vinylimidazole and p-methoxystyrene gave no significant rate enhancement under the same conditions.

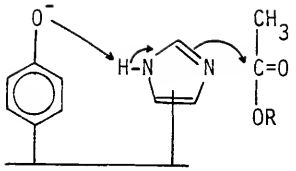


7

This great rate enhancement was attributed to cooperativity between imidazole and phenolate ion, which might involve (i) phenol anion acting as a general base assisting the decomposition of the tetrahedral intermediate and/or (ii) the phenol anion activating a neutral imidazole for nucleophilic attack on the substrate. Cooperative interactions have been demonstrated in small molecules by Bender et al.[7b]



i



ii

Polymer Configuration

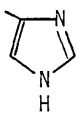
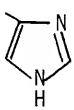
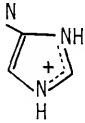
Catalytic properties of polymers are influenced to a large extent by the configuration (conformation) of the polymer in solution. Vinyl polymers are rather flexible as compared with enzymes, i.e., they usually lack specific secondary and unique tertiary structure. As a result, synthetic polymers lack the specific binding pocket which is typical of enzymes. Therefore, the catalytic efficiency of synzymes will depend to a large extent on the pH, ionic strength and composition of the medium, distance of the catalytic group from the polymer backbone, degree of dissociation of catalytic groups, and many other considerations.

Choice of Catalytic Functional Groups

As we have seen above, combinations of cooperative and/or complementary functional groups are necessary to achieve high catalytic efficiency. Catalysis by hydrolytic enzymes is of the nucleophilic and acid-base type. Table I contains a list of functional groups which are directly involved in the catalytic action of some hydrolytic enzymes.

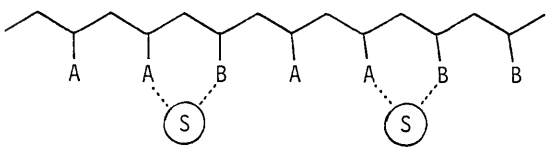
TABLE I

Functional Groups Involved in the Catalytic Action of Some Hydrolytic Enzymes[2a]

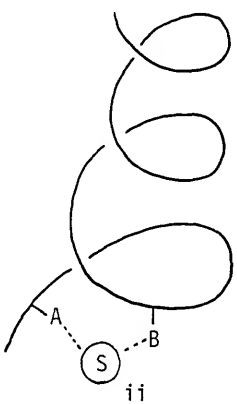
Enzyme	Functional Group
Serine protease	
Chymotrypsin Trypsin α-Lytic protease Elastase Subtilisin	-OH(Ser),  (His), -COOH (Asp)
Papain	-SH(Cys),  (His)
Ribonuclease	 (His),  NH (His, protonated)
Lysozyme	-COOH (Glu), $-COO^-$ (Asp)
Carboxypeptidase	 OH (Tyr), Zn^{2+}

Proposal of Research

As was stated previously, synthetic vinyl macromolecules containing bi- or multi-functionalities have been studied in other laboratories and have shown enzyme-like catalytic activity. These studies have shown a cooperativity between the functionalities leading to a rate enhancement for esterolysis reactions. However, up to this time, functionalities have been introduced into copolymers in random fashion. This ensures a degree of cooperation between the functionalities which is dependent on the degree of alternation (i) or upon the conformation of the polymer chain (ii) as depicted below.



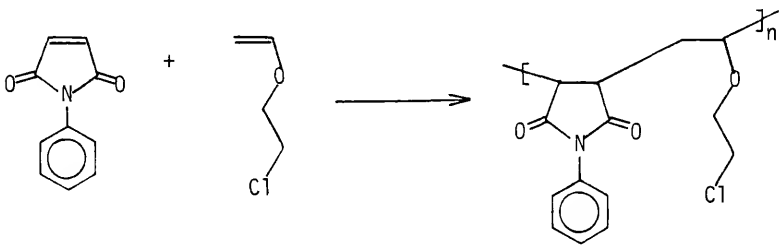
i



ii

It appeared to us that the degree of cooperativity between functional groups could be maximized by preparing regular alternating copolymers. This would assure that each functional group of a given type would be flanked on either side by a functional group of the complementary type.

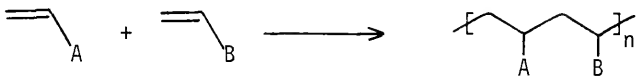
Fundamental to this proposed research is the selection of monomer pairs which undergo regular alternating copolymerization under free-radical conditions. This type of copolymerization is generally thought to result from the formation of a 1:1 charge-transfer complex between electron-rich (donor) and electron-deficient (acceptor) monomer pairs. Some examples of monomer pairs which give regularly alternating copolymers include styrene -- N-phenylmaleimide,[8] 2-chloroethyl vinyl ether -- maleic anhydride,[9] styrene -- maleic anhydride,[10] 2-allyl phenol -- maleic anhydride,[11] and 2-allylphenol -- N-phenylmaleimide.[11] In these laboratories, the alternating copolymerization of N-phenylmaleimide and 2-chloroethylvinyl ether has been studied extensively by Olson.[12]



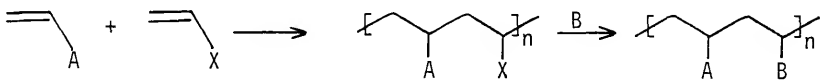
It was concluded in this study that the predominant propagation mechanism is homopolymerization of a 1:1 donor-acceptor complex. An excellent review of the role of the charge-transfer complex in alternating copolymerization can be found in this work.[12]

Our goal of producing bifunctional synzymes via alternating copolymerization could be approached by at least two methods. The first method, which we believed would result in fully functionalized copolymer, is the direct polymerization of monomer pairs containing the

desired functional groups (or the desired functional groups in masked or protected forms).



The second method involves derivatization of a pre-existing alternating copolymer. This method suffers from the fact that polymers can be difficult to functionalize completely and resultant difficulties associated with characterization of a partially functionalized polymer.



We have approached the problem via both methods. Our initial efforts were aimed at direct polymerization of appropriately substituted monomer pairs. Difficulty was encountered effecting copolymerization due to side reactions caused by one of the unprotected functionalities. Hence, derivatization of a pre-existing alternating copolymer was also attempted.

CHAPTER II

EXPERIMENTAL

General

Melting points were determined on a Thomas-Hoover Capillary Melting Point Apparatus or a Fisher-Johns Melting Point Apparatus and are given in degrees celsius (uncorrected). Pressures are expressed in millimeters (mm) of mercury. Elemental analyses were performed by Atlantic Microlabs, Inc., Atlanta, Georgia, and Schwarzkopf Microanalytical Laboratories, Inc., Woodside, New York.

Proton nuclear magnetic resonance (NMR) spectra (60 MHz) were recorded on Varian A-60A or Jeol JNM-PMX-60 instruments. Carbon-13 NMR (25 MHz) and 100 MHz proton NMR spectra were recorded on a Jeol-JNM-FX-100 spectrometer. Chemical shifts are expressed in parts per million (ppm) on the δ scale downfield from tetramethylsilane (TMS) or sodium 2,2-dimethyl-2-silapentane-5-sulfonate (DSS) unless otherwise indicated. In cases where no internal reference was added, spectra were calibrated via a characteristic signal of the deuterated solvent used.[13] The solvent used and calibration information are given in parentheses for each spectrum reported. Multiplicities of proton and off-resonance decoupled carbon resonances are designated as singlet (s), doublet (d), triplet (t), quartet (q), multiplet (m), or broad (br).

Infrared (IR) spectra were recorded on a Perkin-Elmer 281 spectrophotometer. Absorbances are expressed in wavenumbers (cm^{-1}) using polystyrene (1601 cm^{-1}) calibration. Solid samples were run as a KBr pellet; liquid samples were analyzed neat as a thin film between NaCl plates. Absorption bands are assigned the classifications: weak (w), medium (m), strong (s), very strong (vs), broad (br), and shoulder (sh).

Low resolution mass spectra (LRMS) and high resolution mass spectra (HRMS) were recorded on an Associated Electronic Industries (AEI) Model MS-30 spectrometer.

Number average molecular weights ($\overline{M}_n$) of polymers were determined by vapor pressure osmometry (VPO) on a Wescan 233 Molecular Weight Apparatus. Benzil was used as a calibration standard.

Intrinsic viscosities were measured with a Ubbelohde viscometer (dilution viscometer).

Gel Permeation Chromatography (GPC) of polymers was carried out on a Waters Associates - Liquid Chromatograph using glycerated porous glass columns and both ultraviolet (UV) and differential refractometer detectors.

Compound headings appear with the common name(s) listed first, followed by the systematic name as found in Chemical Abstracts (CA). CA registry numbers of known compounds are given in brackets.

Solvents

Deuterated NMR solvents were obtained from the Aldrich Chemical Co. and Merck and Co., Inc. All solvents used for general applications were of Reagent grade or ACS grade quality. For special applications, solvents were distilled. Reference to a distilled solvent

in this chapter indicates that the solvent was purified in the manner described below.[14] Methanol was distilled from $Mg(OCH_3)_2$. Tetrahydrofuran (THF) was distilled from CaH_2. Dimethylsulfoxide (DMSO) and N,N-dimethylformamide (DMF) were allowed to stand over KOH and were distilled from CaO. Dioxane was refluxed with aqueous HCl, dried over KOH and distilled from sodium metal. Ethanol-free chloroform ($CHCl_3$) was obtained by extraction of reagent grade $CHCl_3$ with conc. H_2SO_4 and water, followed by distillation from P_4H_{10}. Dichloromethane (CH_2Cl_2) was distilled from P_4H_{10}. Acetone was distilled from NaI.

Reagents

Starting materials and reagents were obtained from the following suppliers: Aldrich Chemical Co., Fisher Scientific Co., Mallinckrodt, Inc., Eastman Kodak Co., and Polysciences, Inc.

Maleimide and Maleamic Acid Synthesis

3,6-Endoxo-1,2,3,6-tetrahydrophthalic Anhydride/3a,4,7,7a-Tetrahydro-4,7-epoxyisobenzofuran-1,3-dione [5426-0905] (8)

The following procedure was modified from the procedure of Narita et al.[15] To a 1 L three-necked round-bottomed flask equipped with a mechanical stirrer and reflux condenser was added 109.2 g (1.604 mol) of freshly distilled furan and 200 mL of benzene. The solution was cooled via an external ice bath to 0-5°C, after which 157.3 g (1.604 mol) of maleic anhydride was added portionwise. The ice bath was removed after refluxing had slowed, and the mixture was stirred at room temperature for 24 h. Additional benzene was added to facilitate stirring. The mixture was filtered and dried in vacuo to give 235.9 g (88.5%) of white crystalline product (8), mp 115-116°C (dec) [literature mp 118°C (dec)].[16]

^{1}H NMR (DMSO-d_6, TMS): 3.32 (s, 2H), 5.35 (s, 2H), 6.57 (s, 2H).

^{13}C NMR (DMSO-d_6, 39.5): 49.10, 81.75, 136.90, 171.53.

IR (KBr): 3195 (w), 3162 (w), 3136 (w), 3102 (w), 2995 (w), 1860 (s), 1787 (vs, br), 1640 (w), 1595 (w), 1567 (w), 1380 (w), 1321 (m, sh), 1310 (m), 1282 (m), 1242 (m, sh), 1230 (s), 1218 (s), 1193 (m), 1145 (m), 1086 (s), 1020 (s), 1000 (m), 950 (s), 920 (s), 903 (s), 878 (s), 848 (s), 820 (m), 798 (m), 731 (m), 689 (m), 672 (m), 633 (m), 620 (w).

N-Hydroxy-3,6-epoxy-1,2,3,6-tetrahydrophthalimide/3a,4,7,7a-Tetrahydro-2-hydroxy-4,7-epoxy-1H-isoindole-1,3(2H)-dione [5596-17-8] (9)

The following procedure was obtained from Narita et al.[15] To a 1 L round-bottomed flask equipped with a mechanical stirrer and a 250 mL addition funnel was added 75.3 g (1.083 mol) of hydroxylamine hydrochloride (dried in a vacuum oven at 60°C overnight) and 400 mL of freshly distilled methanol. After dissolution, the flask was cooled to 0-5°C. The addition funnel was charged with a solution of 60.8 g (1.083 mol) of potassium hydroxide in 150 mL of freshly distilled methanol and the solution added dropwise with vigorous stirring. After addition, the mixture was stirred an additional 0.5 h, and subsequently suction-filtered into a second 1 L three-necked round-bottomed flask fitted with a mechanical stirrer and reflux condenser. To the stirring hydroxylamine solution was added portionwise 180 g (1.083 mol) of 3,6-endoxo-1,2,3,6-tetrahydrophthalic anhydride (8). The mixture was refluxed for 8 h and then allowed to stir for 13 h at room temperature. The flask was cooled in an ice bath, and

the precipitate was filtered and dried in vacuo to yield 128.6 g (65.5%) of white solid (9), mp 189-195°C (dec) [literature mp 187-188°C (dec)].[15]

^{1}H NMR (DMSO-d_6, TMS): 2.86 (s, 2H), 5.13 (s, 2H), 6.53 (s, 2H).

^{13}C NMR (DMSO-d_6, 39.5): 43.91, 80.00, 136.24, 172.50.

IR (KBr): 3300 (s, br), 3095 (w), 3085 (w), 3050 (w), 3022 (m), 2998 (m), 1787 (s), 1730 (vs, br), 1567 (w), 1437 (s), 1338 (w), 1305 (m), 1283 (m), 1264 (m), 1240 (s), 1227 (m, sh), 1207 (m), 1196 (m), 1150 (s), 1089 (s), 1070 (m), 1010 (m), 951 (m), 916 (m), 881 (s), 846 (m), 823 (m), 803 (m), 792 (m), 720 (s), 644 (s).

N-Acetoxy-3,6-epoxy-1,2,3,6-tetrahydrophthalimide/2-(Acetyloxy)-3a,4,7,7a-tetrahydro-4,7-epoxy-1H-isoindole-1,3(2H)-dione [32463-66-4] (10)

To a 250 mL three-necked round-bottomed flask fitted with a mechanical stirrer and reflux condenser was added 47.7 g (0.263 mol) of 9 and 135 mL of acetic anhydride. The stirred mixture was heated to 89-90°C via an oil bath and maintained at this temperature for 3 h. The resulting solution was cooled to room temperature and placed in a refrigerator overnight. The precipitate was filtered and washed with cold benzene. A second crop of crystals was obtained by concentration of the combined mother liquors in vacuo, followed by precipitation into water. The combined products were recrystallized from benzene and dried in vacuo to afford 34.14 g of white crystals. An additional 10.17 g fraction was obtained from concentration of the benzene mother liquor (75.4%), mp 139-143°C (dec) [literature mp 137-138°C (dec)].[15]

^{1}H NMR ($CDCl_3$, TMS): 2.30 (s, 3H), 2.87 (s, 2H), 5.27 (s, 2H), 6.47 (s, 2H).

^{13}C NMR (DMSO-d_6, 39.5): 17.08, 44.03, 79.85, 136.02, 165.46, 169.26.

IR (KBr): 3095 (w), 3020 (w, sh), 2998 (m), 1810 (s), 1783 (s), 1735 (s), 1430 (w), 1375 (s), 1350 (m), 1284 (m), 1272 (m), 1230 (s), 1218 (s, sh), 1192 (s), 1168 (s), 1139 (s), 1092 (s), 1060 (s), 1012 (m), 992 (m), 985 (m), 915 (m), 878 (s), 845 (m, sh), 836 (m), 817 (m), 800 (s), 739 (m), 710 (m), 693 (m), 637 (m), 629 (m).

N-Acetoxymaleimide/1-(Acetyloxy)-1H-pyrrole-2,5-dione (11)

To a 100 mL one-necked round-bottomed flask was added 49.47 g of 10 and a Telfon-coated stir bar. A short path vacuum distillation head was fitted to the flask, and the pressure of the system was reduced to 25 mm. The flask was heated slowly to approximately 180°C. Decomposition of the solid occurred before 140°C and was accompanied by the evolution of furan. The product was then distilled, bp 140-146°C (24 mm Hg), and collected by cooling the receiving flask. Two recrystallizations from CCl_4 gave 28.36 g (82.5%) of white crystalline product (11), mp 70.5-71.5°C (literature mp 70.5-71.5°C).[15]

^{1}H NMR ($CDCl_3$, TMS): 2.32 (s, 3H), 6.70 (s, 2H).

(DMSO-d_6, TMS): 2.38 (s, 3H), 7.18 (s, 2H).

^{13}C NMR ($CDCl_3$, TMS): 17.33, 132.44, 164.22, 166.80.

(DMSO-d_6, 39.5): 17.03, 133.08, 165.05, 167.49.

IR (KBr): 3080 (w), 3057 (m), 3048 (m), 3003 (w), 2950 (w), 2880 (w), 1818 (s), 1782 (s), 1740 (vs), 1577 (w), 1432 (w), 1380 (s), 1341 (w), 1317 (w), 1177 (s), 1123 (s), 1048 (s), 1007 (w), 820 (s), 778 (m), 670 (s).

Elemental Analysis: Calcd. for $C_6H_5NO_3$: C, 46.46; H, 3.25; N, 9.03. Found: C, 46.49; H, 3.28, N, 9.04.

Phenyl N-(3,6-Epoxy-1,2,3,6-tetrahydrophthalimidyl) Carbonate/3a,4,7,7a-Tetrahydro-2-[(phenoxycarbonyl)oxy]-4,7-epoxy-1H-isoindole-1,3(2H)-dione [60361-88-8] (12)

The following procedure was adopted from Akiyama et al.[17] To a 500 mL three-necked round-bottomed flask fitted with a mechanical stirrer and addition funnel was added 76.4 g (0.422 mol) of 9 and 210 mL of freshly distilled DMF. The flask was cooled to 0-5°C via an external ice bath, and 58.8 mL (0.422 mol) of dry triethylamine was added. The addition funnel was charged with 66.03 g (0.422 mol) of phenyl chloroformate which was added to the stirred solution over a 1 h period. The ice bath was removed and the mixture allowed to stir an additional 3 h. Triethylamine hydrochloride was filtered out and the filtrate precipitated into 2 L of water. Additional product was obtained by dissolving the filtered $Et_3N{\cdot}HCl$ in 1 L of water. Both precipitates were suction filtered and dried in vacuo giving 126.3 g of crude product. Recrystallization from isopropanol afforded 71.4 g (56.3%) of white needles, mp 137-139°C (literature mp 135-136°C).[17]

^1H NMR ($CDCl_3$, TMS): 2.88 (s, 2H), 5.32 (s, 2H), 6.48 (s, 2H), 7.20-7.45 (m, 5H).

^{13}C NMR ($CDCl_3$, TMS): 44.25, 80.47, 120.28, 126.96, 129.74, 136.22, 149.97, 150.70, 168.29.

IR (KBr): 3095 (w), 3075 (w), 3060 (m), 3022 (w), 3003 (w), 1817 (s), 1790 (s), 1740 (vs), 1600 (w), 1585 (m), 1485 (m), 1460 (m), 1360 (m), 1310 (m), 1272 (s), 1230 (vs, br), 1148 (s), 1092 (s), 1068

(s), 1019 (m), 1005 (m, sh), 998 (m), 973 (m), 954 (m), 917 (m), 882 (s), 850 (m), 817 (m), 803 (m), 788 (m), 774 (m), 753 (m), 730 (m), 713 (m), 686 (m), 660 (w), 629 (m), 622 (m).

Phenyl N-Maleimidyl Carbonate/1-[(Phenoxycarbonyl)oxy]-1H-pyrrole-2,5-dione [60361-89-9] (13)

Into a 250 mL Erlenmeyer flask was placed 38.68 g (0.128 mol) of 12, 0.428 g (2.57 mmol) of 4-tert-butylcatechol, 70 mL of bromobenzene, and a few boiling chips. The mixture was heated on a hot plate at such a rate as to allow bromobenzene vapors to condense in the neck of the flask (~160°C inside the flask) for 1.5 h. Bromobenzene was then removed in vacuo and the resulting solid recrystallized from cyclohexane and dried in vacuo giving 24.51 g (81.8%) of maleimide (13) as a pale-yellow solid, mp 99-102.5°C (literature mp 98-99°C).[17] A quantity of 13 was sublimed at 1 mm (100°C) affording white crystals, mp 101-104°C.

^{1}H NMR ($CDCl_3$, TMS): 6.77 (s, 2H), 7.20-7.45 (m, 5H).

^{13}C NMR ($CDCl_3$, TMS): 120.33, 127.06, 129.84, 132.62, 150.75, 163.47.

IR (KBr): 3160 (w), 3100 (m), 3070 (w), 1818 (s), 1783 (s), 1740 (vs), 1587 (m), 1578 (m), 1492 (m), 1482 (m), 1456 (w), 1375 (m), 1220 (vs), 1162 (m), 1155 (m), 1126 (s), 1050 (m), 1023 (m), 1004 (m), 974 (m), 910 (w), 860 (w), 815 (s), 778 (w), 765 (m), 752 (m), 743 (m), 715 (m), 685 (m), 667 (s), 638 (m).

N-Hydroxymaleimide/1-Hydroxy-1H-pyrrole-2,5-dione [4814-74-8] (14)

N-Hydroxymaleimide was prepared via the procedure of Akiyama et al.[17] Thus, to a 100 mL three-necked round-bottomed flask fitted with a reflux condenser and magnetic stir bar was added 7.358 g

(0.0316 mol) of 13 and 40 mL of freshly distilled methanol. The solution was refluxed for 2 h, after which the methanol was removed in vacuo. The residual oil was triturated with a solution of 7:5 hexanes: benzene, and the resulting solid was recrystallized from toluene and dried in vacuo to give 1.55 g (43.4%) of an off-white crystalline solid (14), mp 126-130°C (literature mp 125-126°C).[17] Spectral properties were in agreement with those of an authentic sample kindly supplied by Dr. M. Akiyama.

^{1}H NMR (acetone-d_6, TMS): 6.76 (s, 2H), 9.20 (br, 1H).

(DMSO-d_6, TMS): 6.92 (s, 2H), 10.29 (br, 1H).

^{13}C NMR (acetone-d_6, TMS): 125.50, 160.05.

(DMSO-d_6, 39.5): 131.91, 167.00.

IR (KBr): 3150 (m, br), 3100 (m), 2950 (w), 2850 (w), 1785 (m), 1730 (vs), 1572 (w), 1488 (m), 1306 (w), 1230 (w), 1175 (s), 1130 (m), 1047 (m), 1041 (m), 822 (s), 773 (w), 735 (m), 670 (s).

N-(4-Carbethoxyphenyl)maleanilic Acid/(Z)-4-[(3-Carboxy-1-oxo-2-propenyl)amino]benzoic acid, 1-ethyl ester [53616-17-4] (15)

To a 500 mL Erlenmeyer flask was added 24.4 g (0.148 mol) of ethyl p-aminobenzoate and 250 mL of chloroform. The stirred solution was cooled in an ice bath, and 14.5 g (0.148 mol) of maleic anhydride was added portionwise. After 1 h, the mixture was warmed to room temperature and stirred overnight. The white precipitate was filtered, washed with $CHCl_3$, and dried in vacuo, affording 38.1 g (98%) of maleanilic acid (15). A portion of the product was recrystallized from CH_3CN, mp 190-192°C.

^{1}H NMR (DMSO-d_6, 2.49): 1.28 (t, 3H), 4.25 (q, 2H), 6.41 (AB q, 2H), 7.84 (AB q, 4H), 10.62 (s, 1H).

^{13}C NMR (DMSO-d_6, 39.5): 14.20, 60.51, 118.89, 124.74, 130.30, 131.71, 143.07, 163.78, 165.34, 166.95.

IR (KBr): 3300 (m), 3205 (m), 3110 (m), 2975 (w), 1710 (s), 1635 (m), 1610 (m), 1580 (s), 1540 (s), 1470 (m), 1415 (w), 1405 (m), 1365 (m), 1330 (m), 1310 (m), 1270 (s), 1225 (w), 1175 (m), 1120 (m), 1105 (m), 1025 (m), 1010 (w), 970 (m), 900 (w), 865 (m), 850 (m), 770 (m), 695 (w), 680 (w), 610 (m).

N-(4-Carbethoxyphenyl)maleimide/[4-(2,5-Dihydro-2,5-dioxo-1H-pyrrol-1-yl)benzoic acid, ethyl ester] [14794-06-1] (16)

To a 500 mL one-necked round-bottomed flask was added 38.1 g (0.145 mol) of 15, 1.2 g (0.015 mol) of anhydrous sodium acetate, and 100 mL of acetic anhydride. A magnetic stir bar was added, and a reflux condenser was fitted. The stirring mixture was brought to 90°C over a 1.0 h period, and then allowed to cool to room temperature. The resulting solution was precipitated into 1.5 L of ice-water and allowed to stir overnight. The yellow solid was collected by filtration, recrystallized from ethanol-water, and dried in vacuo, giving 29.57 g (83.4%) of yellow plates, mp 112-113°C (literature mp 113°C).[18]

^{1}H NMR ($CDCl_3$, TMS): 1.38 (t, 3H), 4.37 (q, 2H), 6.84 (s, 2H), 7.80 (AB q, 4H).

^{13}C NMR ($CDCl_3$, 77.0): 13.94, 60.77, 124.86, 129.00, 129.93, 133.97, 135.10, 165.26, 168.63.

IR (KBr): 3470 (w), 3090 (w), 2995 (w), 2900 (w), 1718 (vs), 1710 (vs), 1603 (m), 1507 (m), 1472 (w), 1442 (w), 1405 (m, sh), 1395

(s), 1382 (s), 1365 (m, sh), 1307 (m, sh), 1282 (s), 1213 (w), 1175 (m), 1142 (m), 1128 (m), 1108 (m), 1068 (w), 1022 (m), 948 (w), 853 (m), 830 (m), 764 (m), 699 (m), 684 (m), 638 (w).

N-Hydroxymaleamic Acid [4296-73-5] (17)

N-Hydroxymaleamic acid (17) was prepared from the addition of hydroxylamine [from 61.5 g (0.885 mol) of hydroxylamine hydrochloride neutralized with one equivalent of sodium methoxide in methanol] to a solution of 86.8 g (0.885 mol) of maleic anhydride in distilled dioxane at 0°C. After warming to room temperature and stirring for 1 h, the product was filtered and dried in vacuo, affording 69.3 g (60%) of 17, mp 126-129°C (dec) [literature mp 122-128°C (dec)].[19]

^{1}H NMR (DMSO-d_6, TMS): 6.30 (s, 2H).

^{13}C NMR (DMSO-d_6; 39.5): 129.22, 132.98, 162.17, 165.93.

IR (KBr): 3500-2600 (br, s), 3180 (s), 1695 (m), 1630 (s), 1540 (br, vs), 1400 (s), 1310 (m), 1230 (br, s), 1080 (m), 1065 (s), 990 (m), 980 (m), 915 (m), 847 (m), 800 (m), 730 (m), 630 (m).

N-Carbethoxymaleimide/2,5-Dihydro-2,5-dioxo-1H-pyrrole-1-carboxylic acid, ethyl ester [55750-49-7] (18)

This maleimide was prepared by the method of Keller and Rudinger[20] in 44% yield, mp 55-57°C (literature mp 58-59°C).[20]

^{1}H NMR ($CDCl_3$, TMS): 1.42 (t, 3H), 4.45 (q, 2H), 6.84 (s, 2H).

IR (KBr): 3180 (w), 3100 (m), 2985 (m), 1795 (s), 1770 (vs), 1710 (m), 1595 (m), 1475 (m), 1445 (m), 1398 (m), 1370 (m), 1330 (s), 1265 (s), 1130 (m), 1102 (m), 1053 (m), 1035 (m), 995 (m), 850 (m), 765 (m), 755 (m), 690 (m), 635 (m).

N-[2-(4-Imidazolyl)ethyl]maleamic Acid/(Z)-4-([2-(1H-Imidazol-4-yl) ethyl]amino)-4-oxo-2-butenoic Acid (19)

In an Erlenmeyer flask was combined 0.437 g (3.93 mmol) of histamine (20), 0.367 g (3.74 mmol) of maleic anhydride and 20 mL of chloroform (ethanol-free). The mixture was stirred for 20 h; the solid filtered and dried in vacuo, affording 0.672 g (86%) of 19, which slowly decomposed above 120°C.

^{1}H NMR (D_2O, DSS): 2.95 (m, 2H), 3.52 (m, 2H), 6.13 (AB q, 2H), 7.26 (s, 1H), 8.53 (s, 1H).

IR (KBr): 3600-2400 (br, m), 3230 (w), 3135 (w), 3060 (w), 1655 (m), 1625 (s), 1570 (br, s), 1450 (w), 1430 (w), 1398 (w), 1365 (w), 1313 (w), 1270 (m), 1208 (w), 1185 (m), 1100 (br, m), 1065 (w), 975 (m), 902 (w), 855 (br, m), 815 (m), 730 (w), 715 (m), 638 (m), 610 (m).

N-(2-Thiazolyl)maleamic Acid/(Z)-4-Oxo-4-(2-thiazolylamino)-2-butenoic Acid [19789-91-4] (21)

Into an Erlenmeyer flask was placed 0.922 g (9.21 mmol) of 2-aminothiazole (recrystallized from cyclohexane), 0.902 g (9.20 mmol) of maleic anhydride and 40 mL of acetone. The mixture was stirred for 45 h at room temperature. The solid was filtered, washed with acetone and dried in vacuo, affording 1.308 g (72%) of yellow powder (21), mp 151-153°C (dec).

^{1}H NMR (DMSO-d_6, 2.49): 6.46 (s, 2H), 7.37 (AB q, 2H).

^{13}C NMR (DMSO-d_6, 39.5): 113.97, 128.20, 132.54, 137.90, 157.64, 162.56, 167.14.

IR (KBr): 3090 (w), 3000-2200 (br, m), 1655 (m), 1618 (m), 1565 (br, s), 1435 (m), 1398 (m), 1322 (m), 1270 (s), 1205 (m), 1172 (m), 1060 (m), 905 (m), 850 (m), 775 (m), 725 (m), 705 (m), 650 (m), 620 (m).

N-(4-Carboxyphenyl)maleanilic Acid/4-[(3-Carboxy-1-oxo-2-propenyl) amino]benzoic Acid [36847-92-4] (22)

This compound was previously synthesized in these laboratories[21] from p-aminobenzoic acid and maleic anhydride, mp 234°C (dec).

^{1}H NMR (DMSO-d_6, TMS): 6.19 (AB q, 2H), 7.72 (AB q, 4H), 10.48 (br, 1H).

IR (KBr): 3350-2010 (br, m), 3310 (m), 3210 (w), 3000 (w), 2840 (br, w), 2665 (w), 2540 (w), 2240 (w), 1705 (s), 1690 (s), 1625 (m), 1580 (s), 1540 (vs), 1420 (m), 1405 (m), 1325 (m), 1310 (m), 1290 (s), 1265 (m), 1220 (w), 1175 (m), 1120 (w), 1012 (w), 970 (m), 940 (w), 900 (w), 860 (m), 845 (m), 770 (m), 690 (m), 670 (m), 608 (m).

4-[(3-Carboxy-1-oxo-2-propenyl)amino]benzeneacetic Acid (23)

This compound was previously synthesized in these laboratories[21] from p-aminophenylacetic acid and maleic anhydride, and was used without further purification.

^{1}H NMR (DMSO-d_6, TMS): 3.57 (s, 2H), 6.41 (AB q, 2H), 7.41 (AB q, 4H), 10.43 (br, 1H).

IR (KBr): 3280 (s), 3190 (w), 3050 (m), 2720 (w), 2620 (w), 2390 (w), 2240 (w), 1715 (s), 1685 (s), 1615 (s), 1570 (s), 1535 (vs), 1510 (s), 1425 (m), 1400 (m), 1320 (m), 1300 (m), 1265 (m), 1220 (m), 1200 (w), 1180 (m), 1050 (m), 980 (m), 925 (m), 900 (m), 860 (m), 840 (m), 812 (m), 790 (m), 775 (m), 720 (m), 670 (w), 630 (m), 610 (m).

N-[2-(4-Imidazolyl)ethyl]-3,6-endoxo-1,2,3,6-tetrahydrophthalic Acid (24)

To a 50 mL Erlenmeyer flask was added 2.505 g (0.0136 mol) of histamine dihydrochloride and 15 mL of water. To the stirred solution was carefully added 2.286 g (0.0272 mol) of $NaHCO_3$.

Into another flask was placed 2.263 g (0.0136 mol) of 8 and 22 mL of acetone. The solution of free-base (20) in water was slowly added to the acetone solution with rapid stirring. Addition of additional acetone (200 mL) was necessary to make the flask contents homogeneous. After stirring 1 h, the liquid phase was decanted off, and the remaining oily precipitate stirred over fresh acetone. The resulting fine white solid was collected and dried in vacuo to give 4.898 g of 24, apparently contaminated by NaCl. The solid gradually decomposed upon heating to 135°C.

^{1}H NMR (D_2O, DSS): 2.73 (s, 2H), 2.67-3.63 (m, 4H), 5.07 (d, 2H), 6.43 (m, 2H), 7.12 (m, 1H), 8.48 (d, 1H).

IR (KBr): 3660-2730 (m, br), 3240 (m), 3120 (m), 1715 (w), 1650 (s), 1625 (s), 1555 (s), 1430 (m), 1395 (s), 1310 (w), 1270 (m), 1245 (w), 1218 (m), 1183 (w), 1167 (w), 1092 (w), 1060 (w), 1028 (w), 1000 (w), 982 (w), 972 (w), 930 (w), 900 (m), 838 (m), 820 (m), 808 (w), 752 (w), 730 (m), 702 (m), 627 (m).

Succinimides and Succinamic Acids

N-[2-(4-Imidazolyl)ethyl]succinamic Acid/4-([2-(1H-Imidazol-4-yl) ethyl]amino)-4-oxo-2-butanoic Acid (25)

To an Erlenmeyer flask containing a solution of 0.248 g (2.48 mmol) of succinic anhydride in 5 mL of acetone was added dropwise a

solution of 0.275 g (2.47 mmol) of histamine in 3.5 mL of water, and the solution was stirred overnight. Absolute ethanol and acetone were added, and the precipitate was collected and dried in vacuo, giving 0.239 g (45.8%) of white solid, mp 159-159.5°C.

^{1}H NMR (DMSO-d_6, 2.49): 2.37 (m, 4H), 2.63 (m, 2H), 3.26 (m, 2H), 6.84 (s, 1H), 7.68 (s, 1H), 7.98 (t, 1H).

^{13}C NMR (DMSO-d_6, 39.5): 26.68, 29.75, 30.44, 38.87, 117.19, 134.00, 134.73, 171.34, 174.41.

IR (KBr): 3240 (w), 3155 (m), 3100 (m), 3000 (m), 2940 (m), 2855 (m), 1635 (vs), 1610 (s), 1570 (s), 1450 (w), 1420 (s), 1352 (s), 1285 (w), 1205 (s), 1140 (m), 1110 (m), 1065 (w), 1035 (w), 975 (w), 940 (w), 905 (w), 865 (m), 820 (m, br), 770 (m), 720 (m), 640 (m).

N-[2-(4-Imidazolyl)ethyl]succinimide/1-[2-(1H-Imidazol-4-yl)ethyl]-2,5-pyrrolidinedione (26)

A 25 mL three-necked round-bottomed flask fitted with a stir bar, condenser, and gas inlet tube was assembled hot and was cooled by flushing the apparatus with Ar. To the cool flask was introduced 0.734 g (7.34 mmol) of succinic anhydride and 2 mL of distilled DMF. To the stirred solution was added via syringe a solution of 0.820 g (7.38 mmol) of histamine (20) in 3 mL of DMF. A white solid formed which dissolved when the mixture was heated. The solution was refluxed for 2.5 h and allowed to cool. DMF was removed in vacuo, leaving a brown solid which was recrystallized from $CHCl_3$, mp 164-165°C.

^{1}H NMR (DMSO-d_6, 2.49): 2.58 (s, 4H), 2.67 (m, 2H), 3.56 (m, 2H), 6.83 (s, 1H), 7.57 (s, 1H), 8.87 (br, 1H).

^{13}C NMR (DMSO-d_6, 39.5): 24.83, 28.00, 38.04, 116.36, 133.86, 134.88, 177.57.

IR (KBr): 3440 (w), 3120 (w), 3080 (w), 3035 (w), 2990 (w), 2940 (2), 2830 (m), 2750 (w), 2640 (m), 1765 (m), 1690 (vs), 1575 (m), 1485 (m), 1450 (m), 1438 (m), 1433 (m), 1405 (s), 1330 (s), 1318 (m), 1289 (m), 1260 (w), 1248 (s), 1230 (m), 1150 (s), 1090 (m), 1055 (m), 1030 (w), 1000 (sh, m), 990 (m), 950 (m), 910 (br, m), 840 (m), 825 (m), 798 (m), 772 (m), 660 (m), 630 (m), 608 (w).

N-Acetoxysuccinimide [14464-29-0] (27)

To a dry 250 mL three-necked round-bottomed flask fitted with a mechanical stirrer, N_2 inlet tube and septum cap was added 3.0 g (0.026 mol) of N-hydroxysuccinimide, 100 mL of anhydrous ether, and 20 mL of distilled THF. Dry pyridine (2.06 g, 0.026 mol) was added under N_2, and the flask was cooled to 0-5°C. To the stirred solution was added dropwise via syringe 1.5 mL (0.0265 mol) of acetyl chloride. The mixture was stirred for 0.5 h at 0° and then at room temperature for 1 h. The flask contents were transferred to a separatory funnel and extracted with 1 N HCl. The organic layer was dried over anhydrous $MgSO_4$, the solvent removed in vacuo, and the residue triturated with hexanes to give white solid. The solid was recrystallized from benzene-hexanes, filtered, and dried in vacuo to afford 1.496 g (36%) of needles (27), mp 131-133.5°C (literature mp 132-133°C).[22]

^1H NMR ($CDCl_3$, TMS): 2.33 (s, 3H), 2.82 (s, 2H).

^{13}C NMR ($CDCl_3$, TMS): 17.50, 25.59, 165.71, 169.41.

IR (KBr): 2990 (w), 2940 (w), 1822 (s), 1795 (s), 1745 (vs), 1430 (m), 1380 (s), 1360 (sh, m), 1298 (w), 1255 (m), 1220 (s), 1170

(s), 1160 (sh, s), 1075 (sh, m), 1060 (s), 1045 (m), 1005 (w), 990 (w), 832 (s), 810 (m), 765 (m), 650 (m).

Vinyl Ethers

N-(β-Vinyloxyethyl)imidazole/1-[2-(Ethenyloxy)ethyl]-1H-imidazole (28)

The general procedure for N-alkylation of imidazole described by Fournari et al.[23] was used. To a nitrogen-flushed 500 mL three-necked round-bottomed flask fitted with a mechanical stirrer, condenser, and addition funnel was added 100 mL of freshly distilled THF and 13.2 g (0.339 mol) of potassium metal. The addition funnel was charged with a solution of 25.37 g (0.373 mol) of imidazole in 150 mL of THF, which was added over a 1.5 h period. Refluxing the rapidly stirred mixture for 2 h consumed all visible potassium. A solution of 40 mL (0.39 mol) of 2-chloroethyl vinyl ether (CEVE) and 20 mL of THF was added over 1 h, and reflux was maintained an additional 17 h. Precipitated KCl was removed by filtration, and the solvent evaporated in vacuo. The resulting oil was dried over CaH_2 and distilled (131.5-134°C, 4.0 mm), affording 31.2 g of pale-yellow oil. ^{1}H NMR indicated the presence of imidazole (~25%). The oil was dissolved in THF and stirred over NaH overnight. The precipitate was filtered, and the filtrate concentrated in vacuo and dried over CaH_2. Short-path distillation gave 21.86 g (46.7%) of pure 28, bp 90-92°C (0.5 mm), as a colorless oil. The product was stored over CaH_2 at room temperature.

^{1}H NMR ($CDCl_3$, TMS): 3.74-4.40 (m, 6H), 6.38 (X of ABX, 1H), 6.98 (m, 2H), 7.47 (s, 1H).

(DMSO-d_6, TMS): 3.83-4.47 (m, 4H), 6.46 (X of ABX, 1H), 6.93 (t, 1H), 7.15 (t, 1H), 7.60 (s, 1H).

^{13}C NMR ($CDCl_3$, 77.0): 44.93, 65.96, 86.48, 118.35, 128.08, 136.36, 149.89.

IR (neat, NaCl): 3115 (m), 2940 (m), 2880 (m), 1620 (br, s), 1506 (s), 1465 (m), 1440 (m), 1365 (s), 1325 (s), 1285 (s), 1230 (s), 1195 (br, s), 1150 (sh, m), 1110 (s), 1092 (sh, s), 1078 (s), 1038 (s), 1028 (sh, m), 990 (m), 963 (sh, s), 952 (s), 915 (m), 906 (s), 820 (br, s), 740 (br, s), 662 (s), 622 (s).

LRMS (m/e, rel. intensity): 138 (M^+, 19.2), 137 (11.9), 109 (19.4), 108 (96.4), 95 (14.0), 94 (10.0), 86 (19.7), 84 (32.7), 82 (20.0), 81 (100).

HRMS: m/e 138.07744 (calcd. for $C_7H_{10}N_2O$ = 138.07931).

Elemental Analysis: Calcd. for $C_7H_{10}N_2O$: C, 60.85; H, 7.29; N, 20.27. Found: C, 60.85; H, 7.32; N, 20.28.

N-(β-Vinyloxyethyl)piperidine/1-[2-(Ethenyloxy)ethyl]-piperidine [702-06-7] (29)

This vinyl ether was synthesized via the method of Goette.[24] Thus, to a 250 mL three-necked round-bottomed flask fitted with a magnetic stir bar, condenser, and addition funnel was added 42.5 g (0.506 mol) of $NaHCO_3$, 25 mL of water and 25 mL (0.253 mol) of piperidine. The addition funnel was charged with 77 mL (0.759 mol) of CEVE and the contents added over a 1.5 h period to the gently refluxing mixture. Reflux was maintained for 17 h after addition was complete. Ether was added to the cool flask, and the organic phase was dried over NaOH for several days. Ether and excess CEVE were removed under reduced pressure. The residue was distilled under vacuum, affording

35.2 g (87.0%) of colorless oil (29), bp 75-76.5°C (10 mm) [literature bp 72.3-73°C (7.5 mm)].[24]

^{1}H NMR ($CDCl_3$, TMS): 1.23-1.88 (m, 4H), 2.22-2.80 (m, 4H), 3.72-4.37 (m, 4H), 6.51 (X of ABX, 1H).

^{13}C NMR ($CDCl_3$, 77.0): 23.73, 25.34, 54.39, 57.26, 64.82, 85.58, 151.18.

IR (neat, NaCl): 3120 (w), 3080 (w), 3050 (w), 2940 (s), 2855 (m), 2780 (m), 2750 (m), 1635 (m), 1610 (s), 1478 (m), 1455 (m), 1442 (m), 1383 (w), 1352 (m), 1320 (s), 1305 (m), 1280 (m), 1262 (m), 1200 (s), 1160 (m), 1126 (m), 1090 (m), 1080 (m), 1040 (m), 1023 (w), 1000 (m), 984 (m), 962 (m), 945 (w), 862 (m), 810 (s), 780 (w), 760 (w), 700 (w).

β-Vinyloxyethyl(imidazol-4ylmethyl)piperidinium Chloride (30)

The method reported by Tonellato[25] was employed. To a 50 mL one-necked round-bottomed flask containing a stir bar was added 1.338 g (8.74 mmol) of 4-(chloromethyl)imidazole hydrochloride (31) and 10 mL of anhydrous methanol. To the stirred solution at room temperature was added 2.771 g (17.8 mmol) of 29 in one portion, and the solution was stirred for 0.5 h. Approximately 1 g of Na_2CO_3 was added, the mixture stirred for 5 min, and filtered. The filtrate was reduced in volume on a rotary evaporator and suction filtered again. Slow addition of the filtrate into ether gave an oily precipitate, which was taken up in 10 mL of anhydrous methanol, again stirred over ~1 g of Na_2CO_3, filtered, and the filtrate reduced in vacuo. The viscous oil was triturated with acetonitrile, suction filtered and reduced in volume. Precipitation into ether gave an oily residue. This process

(treatment with Na_2CO_3, etc.) was repeated 2 additional times to ensure complete removal of 29 as its free base. Final drying of the oily precipitate in vacuo overnight afforded 1.998 g (84%, crude) of hydroscopic solid (30). Proton NMR revealed the presence of ether as a major contaminant.

^1H NMR (D_2O, DSS): 1.50-2.17 (m, 6H), 3.25-3.69 (m, 6H), 4.17-4.53 (m, 4H), 4.63 (s, 2H), 6.60 (X of ABX, 1H), 7.52 (s, 1H), 7.86 (s, 1H).

^{13}C NMR (D_2O, DSS): 22.17, 23.15, 58.87, 59.95, 61.80, 63.94, 91.33, 124.57, 129.11, 140.02, 153.18.

IR (KBr): 3600-2500 (br, s), 1625 (s), 1558 (w), 1495 (sh, m), 1465 (m), 1435 (sh, m), 1370 (m), 1325 (m), 1295 (w), 1195 (s), 1090 (m), 1028 (m), 977 (m), 942 (w), 897 (m), 865 (m), 830 (m), 796 (m), 663 (m), 626 (s).

Imidazole and Histamine Derivatives

Histamine/1H-Imidazole-4-ethanamine [51-45-6] (20)

Free base (20) was prepared from histamine dihydrochloride by three methods.

Method A. The procedure of Tabor and Mosettig[26] was used.

A 2% solution of histamine dihydrochloride in 95% ethanol was slowly trickled through a column of Amberlite IRA-400 ion exchange resin. The percolate was reduced to an oil on a rotary evaporator, and distilled under vacuum, bp 134-135°C (0.075 mm) [literature bp 209-210°C (18 mm)],[27] affording a colorless to pale-yellow viscous oil. Distilled yields were generally poor and the purity and boiling point of the product variable.

^{1}H NMR (DMSO-d_6, TMS): 2.42-3.00 (m, 4H), 4.97 (br, 3H), 6.77 (s, 1H), 7.54 (s, 1H).

Method B. To a 50 mL Erlenmeyer flask was added 5.638 g (0.03063 mol) of histamine dihydrochloride and ~10 mL of H_2O. To the stirred solution was added in small increments 5.146 g (0.06125 mol) of $NaHCO_3$, and the flask was allowed to stand overnight. Solvent was removed in vacuo, and the resultant colorless oil was triturated with absolute ethanol. The precipitate was filtered out, and the filtrate reduced in volume in vacuo. Short-path distillation of the resultant oil afforded 1.597 g (46.9%) of 20 as a viscous oil, bp 140-143°C (1.0 mm). Spectral properties of this material were identical to those of 20 made by Method A.

Method C. To an Erlenmeyer flask containing 6.728 g (0.0365 mol) of histamine dihydrochloride dissolved in 10 mL of H_2O was added 18.25 mL (0.0730 mol) of 4 N NaOH solution. The solution was stirred for 0.5 h, and the solvent was removed in vacuo. The resultant thick oil was triturated with 95% ethanol, filtered, and the filtrate reduced to an oil in vacuo. The oil was distilled in a Kugelrohr apparatus (0.050 mm, ~160°C), affording 3.245 g (80%) of a colorless oil, which crystallized on standing, mp 85-88°C (literature mp 83-84°C).[27]

4-(Hydroxymethyl)imidazole Hydrochloride/1H-Imidazole-4-methanol [32673-41-9] (32)

This material was prepared by the method of Totter and Darby.[28] To a 1 L three-necked round-bottomed flask fitted with a mechanical stirrer and reflux condenser was added 250 mL of benzene, 125 mL of water and 50 mL (0.6 mol) of 37% HCl. The mixture was brought to 80°C

via an oil bath, and 50.0 g (0.153 mol) of 4-(hydroxymethyl)imidazole picrate was added in one portion. Stirring was continued until all the solid had dissolved, at which time heating was discontinued and the flask allowed to cool. The aqueous phase was extracted 5 times with 150 mL portions of benzene, stirred over ~2 g of Norit-A and concentrated in vacuo. Recrystallization from absolute ethanol gave 15.45 g (69.3%) of yellow crystals, mp 105-109°C (literature mp 107-109°C).[28] A second recrystallization from absolute ethanol gave 14.25 g of pale-yellow crystals, mp 107-110°C.

^{1}H NMR (D_2O, DSS): 5.00 (br, 2H), 7.52 (br, 1H), 8.79 (br, 1H).

^{13}C NMR (D_2O, DSS): 56.00, 119.26, 134.95, 136.37.

IR (KBr): 3500-2500 (br, s), 1615 (s), 1520 (w), 1458 (s), 1450 (sh, s), 1420 (s), 1362 (m), 1290 (m), 1258 (m), 1252 (m), 1210 (m), 1140 (s), 1070 (s), 1032 (s), 974 (m), 920 (m), 870 (m), 828 (sh, s), 813 (s), 745 (m), 620 (s).

4-(Chloromethyl)imidazole Hydrochloride/4-(Chloromethyl)-1H-imidazole Hydrochloride [31036-72-3] (31)

The procedure of Turner et al.[29] was employed. To a 100 mL three-necked round-bottomed flask equipped with a mechanical stirrer, condenser, drying tube, and addition funnel was added 14.25 g (0.106 mol) of 32 and 10 mL of benzene. The addition funnel was charged with 11 mL (0.15 mol) of thionyl chloride and 20 mL of benzene, and the solution was added to the rapidly stirred suspension over a 1 h period. The mixture was refluxed for 2 h, and then allowed to stand at room temperature overnight. The solid product was suction filtered, washed with benzene and dried in vacuo. Recrystallization from acetonitrile-

absolute ethanol afforded 11.50 g (71.0%) of off-white crystals, mp 141-144°C (literature mp 144°C).[30]

^{13}C NMR (ethanol-d_1, 17.2): 33.23, 117.94, 130.03, 134.56.

IR (KBr): 3500-2500 (s, br), 3140 (m), 1615 (m), 1460 (m), 1430 (m), 1290 (m), 1265 (w), 1168 (w), 1147 (m), 1076 (w), 1062 (m), 978 (m), 922 (m), 900 (m), 853 (m), 808 (m), 765 (w), 720 (m), 675 (w), 620 (s).

Dichlorobis(1-[2-(ethenyloxy)ethyl]-1H-imidazole-N^3)zinc (33)

The method of Eilbeck et al.[31] was employed. In a 25 mL Erlenmeyer flask was weighed 0.198 g (1.45 mmol) of $ZnCl_2$, and 4 mL of absolute ethanol was added. To the stirred solution was added 0.803 g (5.81 mmol) of 28, the transfer aided by 2 mL of ethanol. The solution was stirred for 1 h, at which time anhydrous ether was added dropwise until the solution became turbid. On standing, a colorless oil precipitated from solution. Enough absolute ethanol was added to redissolve the precipitate, and the flask was allowed to stand for 12 days. Upon addition of ether, a white crystalline mass precipitated from solution. The solid was collected, washed with ether, and dried in vacuo, affording 0.535 g (89%) of 33, mp 78.5-80°C.

IR (KBr): 3115 (m), 3040 (w), 2925 (w), 2875 (w), 1635 (sh, m), 1620 (s), 1525 (m), 1442 (w), 1397 (w), 1365 (w), 1322 (m), 1240 (m), 1232 (m), 1187 (s), 1112 (m), 1097 (s), 1033 (m), 992 (m), 952 (m), 850 (m), 830 (m), 760 (m), 668 (m), 653 (m), 630 (m).

Elemental Analysis: Calcd. for $C_{14}H_{20}N_4O_2 \cdot ZnCl_2$: C, 40.75; H, 4.89; N, 13.58; Cl, 17.18. Found: C, 40.72; H. 4.91; N, 13.51; Cl, 17.09.

Dichlorobis(1-methyl-1H-imidazole-N^3)zinc-(T-4) [23570-24-3] (34)

N-methylimidazole-$ZnCl_2$ complex was prepared in an analogous manner to 33. Thus, the reaction of 0.294 g (2.16 mmol) of $ZnCl_2$ and 1.088 g (13.25 mmol) of N-methylimidazole in 15 mL of 95% ethanol gave, after precipitation with ether, 0.512 g (79%) of fine-white solid (34), mp 205-208°C (literature mp 209°C).[32]

IR (KBr): 3150 (w), 3125 (s), 1695 (w), 1635 (w), 1588 (w), 1535 (m), 1520 (m), 1425 (m), 1287 (m), 1235 (s), 1105 (s), 1092 (s), 1025 (w), 955 (m), 847 (m), 775 (m), 740 (m), 668 (m), 653 (s), 620 (m), 615 (m).

Elemental Analysis: Calcd. for $C_8H_{12}N_4 \cdot ZnCl_2$: C, 31.98; H, 4.02; N, 18.64; Cl, 23.60. Found: C, 32.03; H, 4.07; N, 18.59; Cl, 23.59.

N-[(Ethenyloxy)carbonyl)]-1H-imidazole-4-ethanamine (35) and 7,8-Dihydro-5-oxo-imidazo[1,5-c]pyrimidine (36)

To a 100 mL three-necked round-bottomed flask containing a stir bar and an addition funnel was added 5.522 g (0.030 mol) of histamine dihydrochloride, 10 mL of water, and 13 mL of dioxane. To the stirred mixture was added in portions 7.562 g (0.090 mol) of $NaHCO_3$. The flask was cooled to 0°, and a solution of 3.19 g (0.030 mol) of vinyl chloroformate in 12 mL of dioxane was added over 25 min. The mixture was stirred an additional 0.5 h and then warmed to room temperature. NaCl was filtered, and the filtrate was reduced in volume in vacuo. The resulting oil was chromatographed on a basic alumina column (3:2 $CHCl_3$:methanol eluting solvent). One large fraction was collected; removal of solvent in vacuo gave 2.361 g of yellow oil. ^1H NMR indicated the desired mono acylated product (35) was present. The oil was

rechromatographed on a silica gel column (4% methanol:$CHCl_3$ eluting solvent). Two products were isolated, 35, 0.108 g (2%), mp 88-89°C and 36, 0.260 g (6.3%), mp 219-221°C (dec).

35

^1H NMR ($CDCl_3$, TMS): 2.83 (t, 2H), 3.51 (m, 2H), 4.37-4.79 (AB of ABX, 2H), 5.67 (br, 1H), 6.83 (d, 1H), 7.16 (X of ABX, 1H), 7.57 (d, 1H), 9.19 (br, 1H).

^{13}C NMR (acetone-d_6, 29.8): 28.05, 41.69, 94.13, 116.46, 135.71, 136.87, 143.16, 154.18.

($CDCl_3$, TMS): 27.49, 41.04, 95.28, 115.90, 135.00, 136.12, 142.22, 153.91.

IR (KBr): 3220 (m), 3005 (w), 2965 (w), 2940 (w), 1708 (s), 1647 (m), 1570 (m), 1550 (m), 1485 (w), 1450 (m), 1425 (w), 1362 (w), 1310 (m), 1295 (m), 1277 (m), 1260 (m), 1222 (m), 1190 (w), 1158 (m), 1082 (m), 1040 (m), 983 (m), 974 (m), 952 (m), 860 (m), 820 (m), 785 (m), 764 (w), 735 (w), 700 (w), 617 (m).

LRMS (m/e, rel. intensity): 181 (M^+, 0.1), 138 (5.9), 137 (30.6), 82 (10.2), 81 (100).

HRMS: m/e 181.0828 (calcd. for $C_8H_{11}N_3O_2$ = 181.0851).

Elemental Analysis: Calcd. for $C_8H_{11}N_3O_2$: C, 53.03; H, 6.12; N, 23.19. Found: C, 53.09; H, 6.14; N, 23.20.

36

^1H NMR (DMSO-d_6, 2.49): 2.84 (t, 2H), 3.26-3.42 (m, 2H), 6.77 (d, 1H), 8.04 (d, 1H), 8.17 (br, 1H).

^{13}C NMR (DMSO-d_6, 39.5): 19.27, 38.72, 124.64, 127.33, 134.00, 148.38.

IR (KBr): 3220 (br, m), 3115 (s), 2960 (w), 2940 (w), 2920 (w), 2890 (m), 1735 (sh, s), 1710 (s), 1580 (w), 1473 (m), 1455 (m), 1430 (m), 1410 (s), 1360 (w), 1342 (m), 1327 (m), 1310 (w), 1297 (w), 1258 (w), 1232 (w), 1210 (s), 1180 (m), 1150 (m), 1080 (m), 1060 (m), 1045 (m), 930 (m), 865 (w), 833 (m), 755 (m), 680 (m), 650 (m).

LRMS (m/e, rel. intensity): 138 (2.5), 137 (M^+, 26.5), 81 (100).

HRMS: m/e 137.05836 (calcd. for $C_6H_7N_3O$ = 137.05891).

Elemental Analysis: Calcd. for $C_6H_7N_3O$: C, 52.55; H, 5.14; N, 30.64. Found: C, 52.51; H, 5.17; N, 30.65.

N,1-[(Diethenyloxy)carbonyl]-1H-imidazole-4-ethanamine (37)

To a 200 mL three-necked round-bottomed flask equipped with an addition funnel was added 0.97 g (8.7 mmol) of 20 and 50 mL of ethanol-free $CHCl_3$. The solution was cooled to 0°C, and 1.2 mL (8.6 mmol) of dry triethylamine was added. The addition funnel was charged with 0.88 g (8.3 mmol) of vinyl chloroformate and 50 mL of $CHCl_3$, and the contents were added over a 2.5 h period. The flask was allowed to reach room temperature, and water was added. The organic phase was twice extracted with 50 mL portions of water and dried over anhydrous $MgSO_4$. Removal of solvent in vacuo gave a white solid which was recrystallized from cyclohexane, filtered, and dried in vacuo, affording 0.40 g (38% based on vinyl chloroformate) of 37, mp 99-100°C.

^1H NMR ($CDCl_3$, TMS): 2.79 (t, 2H), 3.55 (m, 2H), 4.36-5.24 (2 AB of 2 ABX, 4H), 5.40 (br, 1H), 7.09-7.39 (2 X of 2 ABX, 2H), 7.26 (d, 1H), 8.13 (d, 1H).

^{13}C NMR ($CDCl_3$, 77.0): 27.73, 39.96, 94.79, 100.35, 113.65, 136.95, 140.85, 141.67, 141.97, 145.77, 153.42.

IR (KBr): 3240 (m), 3145 (w), 3050 (m), 2940 (w), 1775 (s), 1735 (s), 1640 (m), 1587 (m), 1560 (m), 1488 (m), 1453 (w), 1440 (w), 1406 (s), 1370 (m), 1327 (m), 1295 (m), 1265 (s), 1247 (s), 1215 (m), 1197 (m), 1173 (m), 1138 (m), 1108 (m), 1055 (m), 1010 (m), 982 (m), 965 (w), 952 (m), 943 (m), 880 (m), 847 (m), 838 (m), 752 (m), 730 (m), 680 (w), 668 (w).

LRMS (m/e, rel. intensity): 253 (0.3), 252 (0.7), 251 (M^+, 0.8), 210 (2.0), 209 (5.2), 208 (40.0), 207 (8.7), 164 (21.3), 152 (26.9), 151 (38.9), 138 (34.0), 137 (13.0), 95 (24.7), 81 (100).

HRMS: m/e 251.0899 (calcd. for $C_{11}H_{13}N_3O_4$ = 251.0906).

Elemental Analysis: Calcd. for $C_{11}H_{13}N_3O_4$: C, 52.59; H, 5.22; N, 16.73. Found: C, 52.58; H, 5.31; N, 17.21.

4-Allylimidazole/4-(2-Propenyl)-1H-imidazole [50995-98-7] (38)

A 250 mL three-necked round-bottomed flask, mechanical stirring rod, condenser, and addition funnel were assembled while hot and cooled by flushing with N_2. To the flask was added 3.408 g (0.140 mol) of Mg turnings; the addition funnel was charged with 100 mL of freshly distilled THF and 10 mL (0.142 mol) of vinyl bromide. Reaction was initiated by addition of a solution of a drop of ethylene bromide in 10 mL of THF. The vinyl bromide solution was then added at a rate which maintained a gentle reflux. When formation of Grignard reagent was complete, the solution was cooled to 0°C via an external ice bath. To the flask was added 4.289 g (0.0280 mol) of 31 in ~15 equal portions over a 2.5 h period. The rapidly stirred mixture was maintained at 0°C for an additional 0.5 h, then allowed to warm to room temperature and was quenched by careful addition of 20 mL of

saturated NH_4Br. Additional water was added to dissolve the precipitated salts, and the organic layer was separated. The aqueous layer was extracted with 2-150 mL portions of $CHCl_3$, and the combined organic fractions were dried over anhydrous $MgSO_4$. Solvent was removed in vacuo, giving dark-yellow oil. This oil was chromatographed on a column of silica gel using a mixture of $CHCl_3$:CH_3OH (95:5) as eluting solvent. Fractions were combined which gave an $R_f \cong 0.30$ by TLC [silica gel, $CHCl_3$:CH_3OH (95.5)]. Removal of solvent in vacuo afforded 1.372 g (45%) of pale-yellow oil (38) having identical ^1H NMR properties as reported by Begg et al.[33]

^1H NMR ($CDCl_3$, TMS): 3.30-3.45 (m, 2H), 5.00-5.20 (m, 2H), 5.79-6.20 (m, 1H), 6.81 (d, 1H), 7.60 (d, 1H), 11.05 (br, 1H).

^{13}C NMR ($CDCl_3$, TMS): 31.39, 116.14, 117.31, 134.76, 135.25, 135.78.

IR (neat, NaCl): 3500-2300 (br, s), 3080 (m), 3015 (w), 2985 (m), 2850 (br, m), 2740 (w), 2640 (w), 1640 (m), 1588 (m), 1570 (m), 1473 (br, m), 1430 (m), 1323 (w), 1298 (m), 1262 (m), 1230 (m), 1195 (w), 1160 (w), 1105 (m), 1088 (m), 990 (s), 940 (m), 915 (s), 820 (m), 750 (m), 662 (m), 625 (m).

LRMS (m/e, rel. intensity): 109 (6.6), 108 (M^+, 68.4), 107 (100), 82 (20.7), 81 (85.5), 80 (86.2), 54 (26.8), 53 (40.9).

HRMS: m/e 108.06875 (calcd. for $C_6H_8N_2$ = 108.06875).

4-Nitroimidazole/4-Nitro-1H-imidazole [3034-38-6] (39)

This material was prepared in accordance with the method of Stambaugh and Manthei[34] in 31% yield, mp 308-309°C (literature mp 308-310°C).[34]

^1H NMR (DMSO-d_6, 2.49): 7.83 (d, 1H), 8.29 (d, 1H).

Other Monomers

2-Propenylphenol/(E) and (Z)-2-(1-Propenyl)-phenol [6380-21-8] (40)

This monomer was synthesized via the isomerization of 2-allylphenol as reported by Tarbell.[35] The product consisted of both E and Z isomers (87%), bp 115-123°C (17 mm), [literature bp 110-115°C (15-16 mm)].[35]

^{1}H NMR ($CDCl_3$, TMS): 1.69 and 1.86 (2 d of d, 3H), 5.34 (br, 1H), 5.85-7.34 (m, 6H).

^{13}C NMR ($CDCl_3$, 77.0): 14.42, 18.71, 115.11, 115.70, 120.33, 120.86, 123.93, 125.25, 127.20, 127.83, 127.98, 128.47, 129.73, 130.86, 152.15.

IR (neat, NaCl): 3540-3300 (br, s), 3060 (m), 3035 (m), 2960 (m), 2935 (m), 2910 (m), 2875 (w), 1850 (m), 1730 (w), 1655 (w), 1605 (m), 1580 (m), 1495 (s), 1483 (s), 1450 (s), 1330 (br, m), 1282 (m), 1225 (br, s), 1172 (s), 1150 (m), 1105 (m), 1080 (m), 1037 (m), 965 (s), 945 (sh, m), 840 (m), 790 (m), 750 (s), 717 (m), 610 (m).

Isoeugenol/2-Methoxy-4-(1-propenyl)-phenol [97-54-1] (41)

Isoeugenol was obtained from the Aldrich Chemical Co. and was distilled before use, bp 138.5-140.5°C (9 mm), [literature bp 140°C (12 mm)].[27]

^{1}H NMR ($CDCl_3$, TMS): 1.82 (d, 3H), 3.80 (s, 3H), 5.72 (s, 1H), 5.76-6.38 (m, 2H), 6.81 (s, 3H).

^{13}C NMR ($CDCl_3$, TMS): 18.27, 55.80, 108.05, 114.44, 119.31, 123.31, 130.81, 144.80, 146.65.

IR (neat, NaCl): 3540-3400 (s, br), 3015 (m), 2960 (m), 2935 (m), 2910 (m), 2880 (w), 2845 (m), 2730 (w), 1590 (m), 1510 (vs), 1462 (s), 1450 (s), 1423 (s), 1370 (m), 1260 (br, s), 1230 (s), 1205 (s), 1153 (s), 1120 (s), 1030 (s), 960 (m), 920 (w), 905 (w), 855 (m), 820 (m), 802 (m), 783 (m), 755 (w), 732 (w).

trans-Anethole/(E)-1-Methoxy-4-(1-propenyl)-benzene [4180-23-8] (42)

This material was purchased from the Aldrich Chemical Co. and was used without further purification.

^{1}H NMR ($CDCl_3$, TMS): 1.82 (d, 3H), 3.73 (s, 3H), 5.92-6.42 (m, 2H), 7.01 (ABq, 4H).

^{13}C NMR ($CDCl_3$, 77.0): 18.32, 55.12, 113.85, 123.30, 126.86, 130.37, 130.81, 158.64.

IR (neat, NaCl): 3029 (m), 3000 (m), 2955 (m), 2930 (m), 2910 (m), 2880 (w), 2835 (w), 2730 (w), 1650 (w), 1605 (s), 1575 (m), 1505 (vs), 1462 (br, m), 1440 (m), 1415 (w), 1375 (w), 1305 (m), 1280 (m), 1245 (s), 1210 (w), 1175 (m), 1110 (m), 1035 (s), 962 (m), 940 (m), 837 (m), 785 (m), 755 (m), 710 (w).

N-Ethylmaleimide/1-Ethyl-1H-pyrrole-2,5-dione [128-53-0] (43)

This monomer was purchased from the Aldrich Chemical Co. (Gold Lable) and was used without further purification.

Diethylfumarate/(E)-2-Butenedioic acid, diethyl ester [623-91-6] (44)

Diethylfumarate (44) was obtained from the Bordon Chemical Co. and was distilled from CaH_2 under reduced pressure, bp 83.5-84°C (4.3 mm) [literature bp 75°C (5 mm)].[36]

^{1}H NMR ($CDCl_3$, TMS): 1.33 (t, 3H), 4.28 (q, 2H), 6.82 (s, 2H).

Fumaronitrile/(E)-2-Butenedinitrile [764-42-1] (45)

This monomer was purchased from the Aldrich Chemical Co. Recrystallization of 45 from benzene gave white needles, mp 94-96.5°C (literature mp 95-97°C).[37]

^{1}H NMR ($CDCl_3$, TMS): 6.29 (s, 2H).

N-Vinylimidazole/1-Ethenyl-1H-imidazole [1072-63-5] (46)

This monomer was purchased from Polysciences, Inc. and distilled from CaH_2 before use, bp 89-90°C (17 mm).

^{1}H NMR ($CDCl_3$, TMS): 4.77-5.33 (AB of ABX, 2H), 6.89 (X of ABX, 1H), 7.07 (s, 1H), 7.18 (s, 1H), 7.66 (s, 1H).

^{13}C NMR (acetone-d_6, 29.8): 101.15, 116.55, 130.10, 130.30, 137.02.

Maleic Anhydride/2,5-Furandione [108-31-6] (47)

Maleic anhydride was obtained from Fisher Scientific Co. and was sublimed at atmospheric pressure (80°C) prior to use, mp 50.5-53°C (literature mp 52.8°C).[27]

Homopolymers

Poly(N-Acetoxymaleimide) (48)

To a heavy-walled polymerization tube was added 2.021 g (0.0130 mol) of 11, 0.0211 g (0.128 mmol) of AIBN, and 25 mL of freshly distilled CH_2Cl_2. After all the solid had dissolved, the tube was degassed (3 freeze-pump-thaw cycles) and sealed at ~10^{-5} mm. Polymerization was carried out in a constant temperature bath (61°C) for 66 h. The tube was opened and the contents precipitated into ether. The solid was collected, redissolved in dioxane and reprecipitated into ether. The solid was again collected and dried in a vacuum oven (100°C) overnight to afford 1.614 g (80% conversion) of pink powder (48).

^{1}H NMR (DMSO-d_6, TMS): 2.34 (br-s, 3H), 3.45, 4.13 (br, 2H).

^{13}C NMR (CD_3CN-$Cl_2CHCHCl_2$, 60°C, 1.30): 17.43, 42.19, 165.74, 169.05, 170.95.

IR (KBr): 2940 (w), 1820 (s), 1790 (s), 1730 (vs), 1625 (w), 1430 (w), 1373 (m), 1220 (s), 1160 (s), 1055 (m), 1000 (w), 820 (m), 725 (w), 640 (m).

Elemental Analysis: Calcd. for $C_6H_5NO_4$: C, 46.46; H, 3.25; N, 9.03. Found: C, 45.74; H, 3.38; N, 8.88.

VPO (acetone): $\overline{M}_n$ = 3850 g/mol.

Poly(Phenyl N-Maleimidyl Carbonate) (49)

To a heavy-walled polymerization tube was added 3.143 g (0.01348 mol) of 13, 0.0250 g (0.152 mmol) of AIBN, and 6 mL of distilled acetone. The tube was transferred to a high-vacuum line, degassed in the usual manner and sealed at ~10^{-5} mm. The polymerization was carried out in a constant temperature bath (60°C) for 89 h. The tube was opened, and the solution was slowly added dropwise to a beaker of vigorously stirred ether. The precipitate was collected and dried in vacuo giving 2.755 g (87% conversion) of pale-green solid (49).

^{1}H NMR (CD_3CN, 1.93): 3.99 (br, 2H), 7.32 (br, 5H).

^{13}C NMR (CD_3CN, 70°C, 1.30): 42.92, 121.39, 128.50, 131.18, 150.63, 151.95, 169.49.

IR (KBr): 3060 (w), 2940 (w), 1825 (s), 1795 (s), 1735 (vs), 1600 (w), 1588 (m), 1490 (m), 1457 (m), 1375 (m), 1290 (m), 1225 (br, vs), 1160 (m), 1115 (w), 1070 (s), 1020 (m), 1005 (m), 960 (m), 905 (w), 840 (w), 775 (m), 750 (m), 682 (m), 630 (m).

Elemental Analysis: Calcd. for $C_{11}H_7NO_5$: C, 56.66; H, 3.03; N, 6.01.
Found: C, 55.44; H, 3.11; N, 6.17.

Poly(N-Hydroxymaleimide) (50) from 48

To a 50 mL Erlenmeyer flask containing a stir bar was added 1.0 g (0.0144 mol) of hydroxylamine hydrochloride and 20 mL of freshly distilled methanol. To the stirred solution was added 3.9 mL (0.0144 mol) of 3.7M sodium methoxide. After 0.5 h, the mixture was suction filtered. To the filtrate was added a solution of 48 in CD_3CN and $Cl_2CHCHCl_2$ (NMR sample ~150 mg), the transfer aided by rinsing the tube with acetone. The resulting mixture (pink ppt.) was stirred for 48 h, and the solvents were then removed in vacuo. Trituration of the resulting solid with water gave pink solid which was suction filtered and dried in vacuo.

^{13}C NMR (acetone-d_6, 29.80): 42.03, 172.75.

IR (KBr): 3640-2300 (br, m), 3470 (br, m), 2920 (w), 2800 (w), 1785 (m), 1700 (br, vs), 1620 (m), 1470 (br, m), 1385 (w), 1340 (w), 1230 (s), 1120 (m), 1070 (m), 728 (m), 645 (m).

Poly(N-Hydroxymaleimide) (50) from 49

To a 50 mL round-bottomed flask containing a stir bar was added 1.411 g (6.05 mmol of repeat units) of 49 and 20 mL of methanol. A reflux condenser was attached, and the mixture was refluxed for 20 h. The cooled solution was precipitated into 200 mL of benzene-pentane (2:1). The solid was reprecipitated from acetone into ether, filtered, and dried in vacuo, giving 0.605 g (88%) of tan powder (50). The product decomposed above 265°C. The IR spectrum of this material was identical to that of 50 derived from 48.

Elemental Analysis: Calcd. for $C_4H_3NO_3$: C, 42.49; H, 2.67; N, 12.39. Found: C, 43.75; H, 3.40; N, 11.94.

Poly[N-(4-Carbethoxyphenyl)maleimide] (51)

To a heavy-walled polymerization tube was added 3.423 g (0.01396 mol) of 16, 0.0270 g (0.164 mmol) of AIBN, and 10 mL of freshly distilled DMF. When all the solid had dissolved, the tube was degassed (3 freeze-pump-thaw cycles) and sealed at ~10^{-5} mm. Polymerization was carried out in an oil bath (75°C) for 44 h. The tube was opened, and most of the DMF was removed in vacuo. The resulting oil was dissolved in 5 mL of acetone and precipitated into ether. The solid was reprecipitated from acetone into ether, collected, and dried in vacuo to give 2.102 g (61% conversion) of pink solid (51).

^{1}H NMR (acetone-d_6, 50°C, TMS): 1.36 (br, 3H), 4.33, 4.40 (br, 4H), 7.47, 8.08 (br, 4H).

^{13}C NMR (acetone-d_6, 50°C, 29.8): 14.55, 41.74, 45.64, 61.77, 127.47, 130.74, 136.44, 165.83, 175.82.

IR (KBr): 2985 (m), 2940 (w), 2910 (w), 1785 (sh, m), 1715 (vs), 1610 (m), 1510 (m), 1470 (w), 1445 (w), 1415 (sh, m), 1385 (s), 1280 (s), 1185 (s), 1110 (s), 1020 (m), 855 (m), 768 (m), 740 (m), 695 (m), 640 (m).

Elemental Analysis: Calcd. for $C_{13}H_{11}NO_4$: C, 63.67; H, 4.52; N, 5.71. Found: C, 63.13; H, 4.67; N, 6.05.

Poly[N-(β-Vinyloxyethyl)imidazole](52)

All attempts to obtain homopolymer (52) of moderate molecular weight and in good yield were unsuccessful. Low yields of oligomers were generally obtained. Polymerization reaction conditions are described in Chapter III, p. 97.

Copolymers

N-(β-Vinyloxyethyl)imidazole - N-Hydroxymaleimide Alternating Copolymer (53)

To a 100 mL round-bottomed flask was added 2.325 g (0.0168 mol) of 28 and 39.95 mL (0.0168 mol) of 0.4 N HCl. Most of the water was removed on a rotary evaporator at room temperature. The resultant viscous oil was transferred to a heavy-walled polymerization tube aided by a few mL of deionized water. Into a 5 mL volumetric flask was placed 2.223 g (0.0143 mol) of 11, and the flask was diluted to the mark with distilled THF. This solution was added to the polymerization tube (previously cooled to -78°C), and the volumetric flask was rinsed with 3 mL of THF. Finally, 0.0847 g (0.313 mmol) of $K_2S_2O_8$ and 0.1237 g (0.315 mmol) of Fe $(NH_4)_2(SO_4)_2 \cdot 6H_2O$ were added. The final volume of solution was 22 mL. The tube was then degassed on a high-vacuum line (3 freeze-pump-thaw cycles) and sealed at $\sim 10^{-5}$ mm. The tube was placed in a 30.0°C water bath for 91 h. The tube was opened and the contents precipitated into CH_3CN. The acetonitrile was decanted off, and the oily precipitate was taken up in 40 mL of 1 N HCl and dialized (2000 MW retention) against deionized water for several days. The precipitated solid was suction filtered and dried in vacuo to afford 0.844 g (20% conversion) of light-brown solid (53).

^1H NMR: See Appendix, p. 122.

^{13}C NMR: See Chapter III, p. 80.

IR (KBr): 3600-3320 (br, m), 3140 (m), 2940 (w), 1780 (m), 1705 (vs), 1575 (w), 1440 (w), 1400 (w), 1355 (w), 1290 (w), 1230 (s), 1105 (s), 1080 (s), 835 (w), 760 (w), 665 (w), 625 (w).

Elemental Analysis: Calcd. for $C_{11}H_{13}N_3O_4$: C, 52.59; H, 5.21; N, 16.73. Found: C, 49.78; H, 4.97; N, 14.68; S, 0.53.

VPO (DMSO): $\overline{M}_n$ = 488 g/mol.

Intrinsic Viscosity (0.1 N HCl, 30.0°C): [η] = 0.112 dL/g.

Dichlorobis(1-[2-(ethenyloxy)ethyl]-1H-imidazole-N^3) zinc -- N-Acetoxymaleimide Alternating Copolymer (54)

To a heavy-walled polymerization tube was added a solution of 0.327 g (2.40 mmol) of $ZnCl_2$ in 6 mL of distilled THF, followed by a solution of 0.630 g (4.56 mmol) of 28 in THF (3 mL). To this solution was added 0.703 g (4.53 mmol) of 11, 0.0075 g (0.046 mmol) of AIBN, and 6 mL of THF. The solution was degassed on a vacuum line and sealed at ~10^{-5} mm. Polymerization was carried out at 70°C for 3.5 h. The white precipitate was filtered, washed with THF and dried in vacuo. The material was extracted (Soxhelet) with THF for 3 days and dried in vacuo, affording 1.110 g (67.5% conversion) of white solid (54) which decomposed above 220°C.

^{13}C NMR: See Chapter III, p. 89.

IR (KBr): 3640-3340 (br, m), 3135 (m), 2940 (m), 2880 (w), 1818 (s), 1785 (s), 1730 (vs), 1650 (br, w), 1522 (m), 1440 (m), 1370 (m), 1290 (w), 1220 (s), 1165 (s), 1110 (s), 1095 (s), 1065 (m), 950 (m), 830 (m), 755 (m), 655 (m), 625 (w).

Elemental Analysis: Calcd. for $C_{26}H_{30}N_6O_{10} \cdot ZnCl_2$: C, 43.20; H, 4.18; N, 11.63; Cl, 9.81. Found: C, 42.35; H, 4.18; N, 10.98; Cl, 9.22.

Intrinsic Viscosity (DMSO, 30.0°C): [η] = 0.043 dL/g.

N-(β-Vinyloxyethyl)imidazole -- Fumaronitrile Copolymer (55)

To a heavy-walled glass polymerization tube was added 1.878 g (0.0136 mol) of 28, 1.116 g (0.0143 mol) of 45, 0.0466 g (0.284 mmol) of AIBN, and 25 mL of distilled CH_2Cl_2. The tube was transferred to a high-vacuum line, degassed via several freeze-pump-thaw cycles and sealed at ~10^{-5} mm. The polymerization was carried out at 60°C for 44 h, resulting in red solution containing an oily dark precipitate. The tube contents were poured into ether. The oily precipitate was taken up in acetone-methanol and precipitated into chloroform. The solid was reprecipitated from acetone into carbon tetrachloride and dried in vacuo (room temperature) overnight, affording 0.367 g of mustard-brown powder. The mother liquors (from precipitations) were combined and reduced in volume on a rotary evaporator. Precipitation into chloroform gave an additional 0.525 g of dark-brown powder.

IR (KBr): 3150 (m), 2970 (m), 2940 (m), 2250 (m), 2200 (s), 2140 (m), 1620 (s), 1545 (m), 1440 (m), 1420 (m), 1355 (w), 1330 (m), 1290 (m), 1170 (m), 1080 (m), 1035 (w), 830 (m), 750 (m), 665 (m), 625 (m).

Elemental Analysis: Calcd. for $C_{11}H_{12}N_4O$: C, 61.10; H, 5.59; N, 25.91. Found: C, 60.53; H, 4.38; N, 28.62.

N-(β-Vinyloxyethyl)imidazole -- Diethylfumarate Copolymer (56)

To a polymerization tube was added 1.202 g (8.70 mmol) of 28, 1.199 g (6.96 mmol) of 44, 0.0114 g (0.0694 mmol) of AIBN and 25 mL of distilled acetone. The tube contents were degassed (4 freeze-pump-thaw cycles) and the tube sealed at ~10^{-5} mm. Polymerization was carried out at 60°C for 90 h. The acetone solution was

precipitated into cold ether, and the gummy precipitate was dried in a vacuum oven (50°C, 48 h) to give 0.262 g of brittle solid (56).

^{13}C NMR: See Appendix, p. 123.

IR (KBr): 3110 (w), 2980 (m), 2940 (m), 2905 (w), 2870 (w), 1730 (vs), 1595 (m), 1505 (m), 1465 (m), 1445 (m), 1370 (m), 1230 (br, s), 1175 (s), 1160 (s), 1095 (m), 1075 (m), 1025 (s), 905 (w), 855 (m), 815 (w), 740 (m), 660 (m), 620 (w).

Elemental Analysis: Calcd. for $C_{15}H_{22}N_2O_5$: C, 58.05; H, 7.14; N, 9.03. Found: C, 56.82; H, 7.11; N, 6.23.

Isoeugenol -- Maleic Anhydride Copolymer (57)

To a dry heavy-walled polymerization tube was added 2.758 g (0.0168 mol) of 41 and a solution of 1.652 g (0.0168 mol) of 47 and 0.0555 g (0.338 mmol) of AIBN in 10 mL of distilled acetone. The solution immediately turned yellow in color. The tube contents were degassed on a high-vacuum line and the tube sealed at ~10^{-5} mm. Polymerization was carried out at 60°C for 72 h. The viscous acetone solution was added dropwise to a beaker of rigorously stirred CH_2Cl_2, the precipitate filtered and dried in vacuo affording 2.725 g (62% conversion) of white solid (57).

^{13}C NMR: See Appendix, p. 124.

IR (KBr): 3580-3300 (m), 2965 (w), 2940 (w), 1855 (m), 1775 (vs), 1605 (m), 1515 (s), 1460 (m), 1430 (m), 1370 (m), 1275 (s), 1235 (s), 1215 (sh, s), 1155 (m), 1130 (m), 1080 (m), 1030 (m), 920 (s), 825 (m), 785 (w), 735 (w), 645 (w).

Elemental Analysis: Calcd. for $C_{14}H_{14}O_5$: C, 64.12; H, 5.38. Found: C, 63.47; H, 5.61.

VPO (acetone): $\overline{M}_n$ = 6950 g/mol.

Intrinsic Viscosity (acetone, 30.0°C): $[\eta]$ = 0.183 dL/g.

2-Propenylphenol -- Maleic Anhydride Copolymer (58)

To a dry heavy-walled polymerization tube was added 2.331 g (0.0174 mol) of 40 and a solution of 1.703 g (0.0174 mol) of 47 and 0.0542 g (0.330 mmol) of AIBN in 10 mL of distilled acetone. The solution became yellow, and the color persisted throughout polymerization. The tube contents were degassed and the tube sealed at ~10^{-5} mm. Polymerization was carried out at 60°C for 44 h. The viscous acetone solution was added dropwise to a beaker of vigorously stirred CH_2Cl_2. The precipitate was filtered and dried in vacuo, affording 3.510 g (87% conversion) of white solid (58).

^{13}C NMR: See Appendix, p. 125.

IR (KBr): 3660-2500 (m, br), 1855 (m), 1770 (s, br), 1610 (m), 1585 (m), 1485 (m), 1455 (m), 1365 (m, br), 1225 (s), 1150 (s, br), 920 (m, br), 755 (s).

Elemental Analysis: Calcd. for $C_{13}H_{12}O_4$: C, 67.24; H, 5.21. Found: C, 63.94; H, 5.69.

VPO (acetone): $\overline{M}_n$ = 21,200 g/mol.

Intrinsic Viscosity (acetone, 30.0°C): $[\eta]$ = 0.231 dL/g.

Isoeugenol -- N-[2-(4-Imidazolyl)ethyl]maleimide Copolymer (59)

A 25 mL three-necked round-bottomed flask fitted with a stir bar, condenser, and gas inlet tube was assembled hot and cooled via flushing with Ar. To the flask was introduced 0.3463 g (1.32 mmol of repeating units) of 57 and ~2 mL of distilled DMF. To this solution was added 0.1707 g (1.54 mmol) of 20 in 0.5 mL of DMF. A white

precipitate was observed. The mixture was refluxed for 6 h and allowed to cool. The viscous solution was added dropwise to a beaker of rapidly stirred $CHCl_3$; the precipitate was filtered and dried in vacuo. The product was subjected to Soxhelet extraction with $CHCl_3$ for 48 h and dried in vacuo, affording 0.473 g of off-white solid (59). ^{13}C NMR: See Appendix, p. 126.

IR (KBr): 3550-2500 (br, m), 3140 (m), 1765 (m), 1690 (s), 1615 (m), 1595 (m), 1510 (m), 1445 (m), 1400 (m), 1360 (m), 1270 (m), 1225 (br, m), 1160 (m), 1130 (m), 1080 (w), 1025 (m), 900 (br, w), 820 (m), 785 (m), 650 (w), 620 (m).

Elemental Analysis: Calcd. for $C_{19}H_{21}N_3O_4$: C, 64.21; H, 5.96; N, 11.82. Found: C, 58.75; H, 5.66; N, 10.68.

2-Propenylphenol--N-[2-(4-Imidazolyl)ethyl]maleimide Copolymer (60)

Copolymer 60 was prepared from 58 by the same method as copolymer 59 . Thus, 0.383 g (1.65 mmol of repeat units) of 58 was combined with 0.198 g (1.78 mmol) of 20 in refluxing DMF to give 0.470 g of off-white product (60).

IR (KBr): 3650-2500 (br, m), 2960 (m), 1765 (m), 1690 (s), 1590 (m), 1483 (m), 1450 (m), 1400 (m), 1360 (m), 1255 (m), 1220 (m), 1160 (m), 1100 (m), 980 (w), 935 (w), 830 (w), 755 (m), 660 (w), 615 (w).

Elemental Analysis: Calcd. for $C_{18}H_{19}N_3O_3$: C, 66.45; H, 5.89; N, 12.91. Found: C, 62.00; H, 5.56; N, 10.55.

trans-Anethole -- Maleic Anhydride Copolymer (61)

To a polymerization tube was added 1.630 g (0.0166 mol) of 47, 0.0534 g (0.325 mmol) of AIBN, 2.464 g (0.0166 mol) of 42, and 10 mL of distilled acetone. The tube contents were degassed in the usual manner and sealed at ~10^{-5} mm. Polymerization was carried out for 20 h at 60°C. The gelatinous mass was dissolved in DMF and precipitated into ether. The precipitate was filtered and dried in a vacuum oven (90°C, 1 mm) for 48 h, affording 2.065 g (50% conversion) of white solid (61).

^{13}C NMR: See Appendix, p. 127.

IR (KBr): 2960 (m), 2940 (m), 2840 (w), 1860 (m), 1780 (vs), 1610 (m), 1580 (w), 1510 (s), 1465 (m), 1440 (m), 1390 (w), 1335 (m), 1305 (m), 1255 (s), 1180 (s), 1080 (m), 1030 (m), 920 (s), 830 (m), 815 (sh, m), 738 (m).

Elemental Analysis: Calcd. for $C_{14}H_{14}O_4$: C, 68.28; H. 5.73. Found: C, 68.11; H, 5.77.

trans-Anethole -- N-[2-(4-Imidazolyl)ethyl]maleimide Copolymer (62)

Copolymer 62 was prepared from 61 by the same method as copolymers 59 and 60. Thus, 0.422 g (1.71 mmol of repeat units) of 61 was combined with 0.200 g (1.80 mmol) of 20 in refluxing DMF to give 0.402 g of off-white product (62).

IR (KBr): 3440-2800 (br, m), 2940 (m), 2840 (m), 1770 (m), 1695 (s), 1610 (m), 1580 (w), 1510 (s), 1460 (sh, m), 1440 (m), 1400 (m), 1355 (m), 1300 (w), 1250 (m), 1180 (m), 1160 (m), 1105 (w), 1085 (w), 1030 (m), 975 (w), 930 (w), 830 (m), 730 (w), 660 (w), 615 (m).

Isoeugenol -- N-Ethylmaleimide Copolymer (63)

To a polymerization tube was added 1.5315 g (0.0122 mol) of 43, 0.0397 g (0.242 mmol) of AIBN, 2.010 g (0.0122 mol) of 41, and 10 mL of distilled acetone. The tube contents were degassed and the tube sealed at ~10^{-5} mm. Polymerization was carried out at 60°C for 38 h. The acetone solution was added dropwise to a vigorously stirred beaker of ether. The precipitate was filtered and dried in vacuo, affording 3.117 g (88% conversion) of white solid (63).

^{13}C NMR: See Appendix, p. 128.

IR (KBr): 3680-3100 (br, m), 2970 (m), 2940 (m), 2880 (w), 2840 (w), 1770 (m), 1690 (vs), 1600 (m), 1510 (s), 1450 (m), 1405 (s), 1375 (m), 1350 (m), 1270 (m), 1225 (s), 1130 (m), 1030 (m), 940 (w), 895 (w), 860 (w), 810 (m), 790 (m), 770 (w), 730 (w), 650 (m).

Elemental Analysis: Calcd. for $C_{16}H_{19}NO_4$: C, 66.42; H, 6.62; N, 4.84. Found: C, 65.66; H, 6.64; N, 5.03.

VPO (acetone): $\overline{M}_n$ = 25,200.

Intrinsic Viscosity (acetone, 30.0°C): $[\eta]$ = 0.276 dL/g.

2-Propenylphenol -- N-Ethylmaleimide Copolymer (64)

To a polymerization tube was added 1.8594 g (0.01486 mol) of 43, 0.0485 g (0.295 mmol) of AIBN, 1.994 g (0.01486 mol) of 40, and 10 mL of distilled acetone. The tube contents were degassed in the usual manner and the tube sealed (~10^{-5} mm). Polymerization was carried out at 60°C for 38 h. The acetone solution was added dropwise to ethyl ether, the precipitate filtered and dried in vacuo, affording 1.286 g (33% conversion) of white solid (64).

^{13}C NMR: See Appendix, p. 129.

IR (KBr): 3660-3100 (br, m), 2975 (m), 2940 (m), 2880 (w), 1770 (m), 1695 (vs), 1605 (m), 1500 (m), 1485 (m), 1450 (s), 1405 (s), 1378 (m), 1350 (s), 1225 (s), 1135 (m), 1095 (m), 1040 (w), 935 (m), 850 (w), 830 (w), 810 (m), 755 (m), 680 (w).

Elemental Analysis: Calcd. for $C_{15}H_{17}NO_3$: C, 69.48; H, 6.61; N, 5.40. Found: C, 67.01; H, 6.51; N, 6.25.

Intrinsic Viscosity (acetone, 30.0°C): $[\eta]$ = 0.083 dL/g.

Miscellaneous Reactions

Reaction of N-Acetoxymaleimide (11) and N-(β-Vinyloxyethyl)imidazole (28). Preparation of N-Acetoxymaleimide Cyclotrimer (65)

To a 50 mL Erlenmeyer flask was added 4.038 g (0.0260 mol) of 11 and 10 mL of CH_2Cl_2. To this colorless solution was added a solution of 0.0180 g (0.130 mmol) of 28 in 1 mL of CH_2Cl_2. Immediately a red color became apparent which intensified with time. The flask was allowed to stand at room temperature for 165 h. The dark-red solution was added dropwise to a beaker of vigorously stirred ether. The precipitate was filtered and dried in vacuo at 100°C to afford 0.66 g (16.3%) of purple powder (65).

^1H NMR (CD_3CN, TMS): 2.29 (br, ~3H), 3.00-4.60 (br-m, ~2H).

^{13}C NMR: See Appendix, p. 130.

IR (KBr): 2945 (w), 1820 (s), 1790 (s), 1740 (vs), 1430 (w), 1375 (m), 1225 (s), 1160 (s), 1065 (m), 1005 (w), 820 (m), 670 (w), 645 (w).

Elemental Analysis: Calcd. for $(C_6H_5NO_4)_3$: C, 46.46; H, 3.25; N, 9.03. Found: C, 46.40; H, 3.24; N, 9.03.

LRMS (m/e, rel. intensity): 465 (m^+, 0.1), 423 (0.4), 113 (0.4), 60 (19.3), 45 (24.4), 44 (61.8), 43 (100).

VPO (acetone): $\overline{M}_n$ = 488 g/mol

Reaction of Maleic Anhydride (47) and N-(β-Vinyloxyethyl)imidazole (28)

To a 125 mL Erlenmeyer flask was added 0.967 g (7.00 mmol) of 28 and 20 mL of CH_2Cl_2. To this colorless solution was added 0.687 g (7.00 mmol) of 47. The solution immediately turned yellow in color and eventually became brown. The flask was allowed to stand for 11 days. The solution was decanted, leaving a black precipitate which was removed from the flask and stirred with 100 mL of acetone for 3 h. The solid was filtered and dried in vacuo, affording 0.897 g of brown powder.

IR (KBr): 3650-2320 (br, m), 3140 (m), 2950 (w), 1770 (s), 1720 (br, s), 1620 (m), 1580 (m), 1555 (m), 1445 (w), 1380 (br, m), 1220 (br, m), 1190 (m), 1135 (m), 1085 (m), 1035 (m), 935 (m), 830 (m), 750 (m), 665 (w), 625 (m).

Reaction of N-Hydroxymaleimide (14) and N-(β-Vinyloxyethyl)imidazole (28)

To a 50 mL Erlenmeyer flask was added 0.326 g (2.88 mmol) of 14 and 10 mL of distilled acetone. To this pale yellow solution was added 0.406 g (2.94 mmol) of 28. The solution immediately assumed a darker yellow color, and a precipitate began to form. After stirring for 20 h, the precipitate was filtered, washed with acetone and dried in vacuo, affording 0.421 g of yellow solid which decomposed upon heating to 180°C.

IR (KBr): 3140 (w), 3100 (w), 2940 (w), 1785 (m), 1695 (br, s), 1615 (m), 1570 (w), 1555 (w), 1540 (w), 1415 (w), 1360 (w), 1320 (w), 1235 (br, m), 1190 (m), 1080 (m), 955 (w), 830 (w), 740 (w), 690 (m).

Elemental Analysis: Calcd. for $(C_4H_3NO_3)_3 \cdot C_7H_{10}N_2O \cdot H_2O$: C, 46.06; H, 4.27; N, 14.14. Found: C, 45.92; H, 4.21, N, 13.96.

Kinetic Measurements

Equipment and Materials

Pseudo first-order kinetics were measured on Cary 17-D or Perkin-Elmer 330 spectrophotometers. Temperature control was provided by a Lauda K-2/R (40.0 ± 0.2°C) or a Haake A80 (25.0 ± 0.2°C) constant temperature apparatus. A Corning-125 pH meter fitted with a Ag/AgCl pH electrode was used to measure the pH of solutions before and after the reaction with substrate. p-Nitrophenyl acetate (PNPA) was obtained from the Aldrich Chemical Co. and was recrystallized from cyclohexane before use, mp 77-78°C (literature mp 81-82°C).[38] 2,4-Dinitrophenyl benzoate (DNPB) was kindly supplied by Ms. Ann Mobley.[39] Deionized water was distilled in glass before use. DMSO and THF were purified as previously described. Tris(hydroxymethyl)aminomethane (Tris) was obtained from Fisher Chemical Co. and was used without further purification.

Kinetic measurements were carried out under two sets of conditions: Method A for the catalysts imidazole, 50, and 53, and Method B for catalysts 26, 59, 60, 62, 63 and 68.

Buffer Solutions

In Method A, two stock buffer solutions were prepared, 0.1M Tris

and 0.1M Tris·HCl, both having an ionic strength (μ) of 0.1 (KCl). The first solution was prepared by adding 12.114 g (0.100 mol) of Tris and 7.455 g (0.100 mol) of KCl to a 1 L volumetric flask and diluting to the mark with distilled water. Tris·HCl was prepared by adding an ampule of 0.1 N HCl (Acculute), 12.114 g (0.100 mol) of Tris and 7.455 g (0.100 mol) of KCl to a 1 L volumetric flask and diluting to the mark with distilled water. These two solutions were combined to give a buffer solution of the desired pH. Thus a 2:1 volume ratio of Tris·HCl:Tris gave a pH of 7.86 and a 3:1 ratio of Tris·HCl:Tris gave a pH of 7.68 at 25°C.

In Method B, two 80% DMSO:H_2O (v/v) stock solutions were prepared, both 0.02M in Tris or Tris·HCl, μ = 0.02 (KCl). The first solution was prepared by adding 1.2114 g (0.010 mol) of Tris, 0.7455 g (0.010 mol) of KCl, and 100 mL of distilled water to a 500 mL volumetric flask and diluting to the mark with distilled DMSO. The second solution was prepared in the same manner, substituting 100 mL of 0.1N HCl (aq) for 100 mL of distilled water. Again, the solutions were combined to give buffer solution of the desired pH. Thus, a 4:1 volume ratio of Tris·HCl:Tris gave a pH of 8.9.

Substrate Solutions

In Method A, a stock solution of PNPA (2.69×10^{-3}M) in acetonitrile was used. In Method B, stock solutions of PNPA (1.60×10^{-3}M) in DMSO and DNPB (1.60×10^{-3}M) in THF were employed.

Catalyst Solutions

In both Methods A and B, catalyst solutions were prepared according to the concentration of functional groups. Due to difficulty in

determining the exact composition of copolymers, it was assumed that the copolymers studied were strictly 1:1 alternating copolymers. The contribution to the molecular weight by endgroups was also neglected in all polymer catalysts. Thus, stock solutions of all copolymer catalysts studied in Method B were ~5.33 x 10^{-3}N in repeat units dissolved in 0.02M Tris buffer of the desired pH.

Kinetic Method

The following paragraph describes the kinetic method using the Cary 17-D spectrophotometer. The same procedure was used in conjunction with the Perkin-Elmer 330 spectrophotometer with the exception that each sample cell required a corresponding reference cell. To each of 6 -one cm path length quartz cells was added 3.0 mL of buffer solution via pipet. To 4 of the cuvettes was added 150 µl of catalyst solution via micropipet; to the other 2 cuvettes was added 150 µl of buffer solution. To one of the latter cuvettes was added 50 µl of substrate solvent (CH_3CN, DMSO, or THF), and it was placed in the reference beam of the spectrophotometer. The remaining 5 cuvettes were placed in the sample compartment to equilibrate thermally. The sample cuvettes were then each charged with 50 µl of substrate solution, agitated by inverting the sample holder, and replaced in the sample compartment. The release of p-nitrophenolate ion (Method A - 400 nm, Method B - 416 nm) or 2,4-dinitrophenolate ion (Method B - 370 nm) was observed at constant wavelength at constant time intervals.

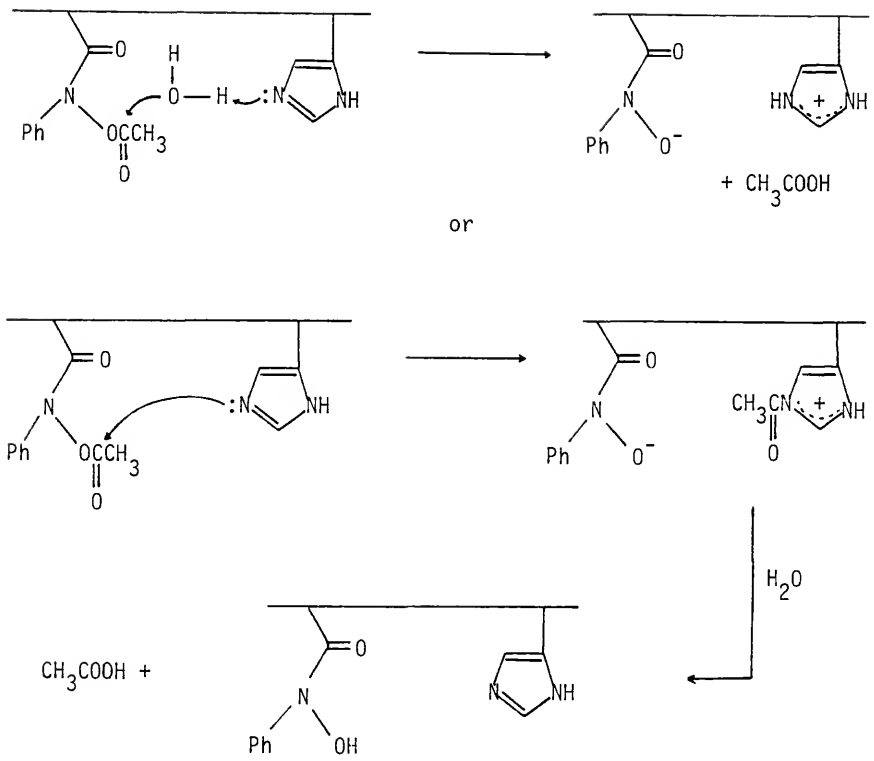
The reaction was followed for at least 10 half-lives as judged by the constancy of the absorbance readings (A_∞). A plot of ln (A_∞-A_t) vs time (t) was constructed, and the negative slope of the best

straight line as determined by the least squares program of a Texas Instruments TI-55-II calculator gave the desired rate constants (k_{meas}). As k_{meas} is the sum of the catalyzed (k_{obs}) and uncatalyzed (k_{blank}) rate constants, it was necessary to subtract k_{blank} from k_{meas} to obtain k_{obs}. Furthermore, the second-order rate constant (k_{cat}) was calculated from the relation $k_{cat} = k_{obs}/[catalyst]$.[40] In the case of slow reactions where A_{∞} was not obtained in a reasonable time, k_{meas} was determined by the method of Kezdy and Swinbourne, which is described in a monograph by Espenson.[41]

CHAPTER III

RESULTS AND DISCUSSION

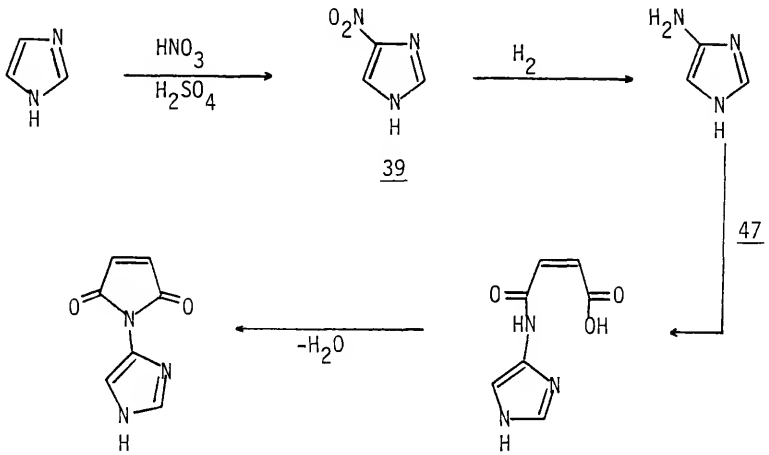
As was stated in Chapter I, alternating copolymers containing pendant groups which would exhibit cooperative behavior in the hydrolysis of an ester substrate were sought. It was decided to utilize substituted vinyl ether and maleimide monomer pairs in order to achieve the desired alternation of pendant functional groups, as this combination of monomers is known to give regularly alternating copolymers under free-radical initiation conditions.[12] The selection of catalytically active functional groups was made possible by the work of Kunitake et al.[6,42-44] In these studies, it was shown that the hydroxamic acid group is an excellent acylation catalyst for activated ester substrates. However, decomposition of an acylhydroxamate is a slow process. In order to obtain a useful catalyst, i.e., one with efficient turnover of the catalytic group, the deacylation rate must be comparable to the acylation rate. Kunitake and Okahata[6] found that introduction of an imidazole group into the polymer will accelerate the deacylation process. It was concluded that the imidazole group assists deacylation of the acylhydroxamate intermediate either by acting as a general base or as a nucleophilic catalyst as depicted below.



With this information in mind, the synthesis of maleimide and vinyl ether monomers substituted with imidazole and hydroxamic acid functionalities was initiated. Initial effort was directed at attaching an imidazole group to a maleimide; however, due to the base sensitive nature of maleimides,[45] it was decided that the hydroxamic acid-maleimide combination would be more compatable. This logic dictated that the complementary vinyl ether monomer should contain an imidazole group. In the next section are described several stratagies to carry out this objective.

Imidazole --Maleimides

Our initial attempt to prepare an imidazole - maleimide is outlined schematically below.



Imidazole was nitrated via a literature procedure[34] to afford 4-nitroimidazole (39). We envisioned obtaining the desired N-(4-imidazolyl) maleimide via reaction of 4-aminoimidazole with maleic anhydride (47) followed by dehydration of the resulting maleamic acid. This attempt was thwarted by the inability to obtain 4-aminoimidazole from reduction of 39. Indeed, 4-aminoimidazole is very unstable and has been isolated only as dihydrochloride and sesquipicrate salts.[46] Our attempts to convert 39 to 4-aminoimidazole are outlined in Table II.

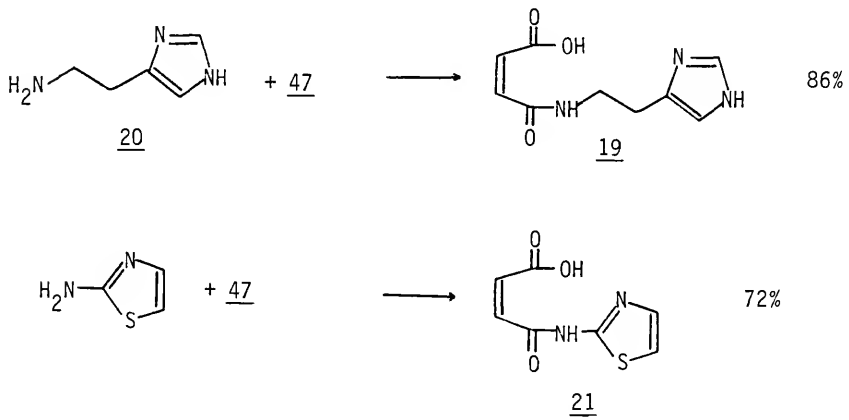
In lieu of 4-aminoimidazole, the reactions of histamine (20) and 2-aminothiazole with maleic anhydride were carried out. Although the maleamic acids were obtained in reasonable yield, attempts to effect

TABLE II

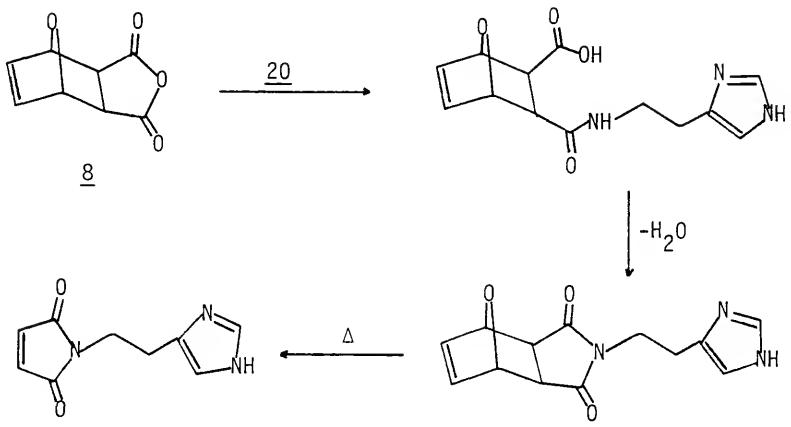
Hydrogenation of 4-Nitroimidazole (39)

Reaction Conditions	Work Up	Comments
H_2/5% Pd on C DMSO/1 atm.	DMSO solution diluted with ether, treated with anhy. HCl	black tar obtained
H_2/10% Pd on C DMSO/~4 atm.	DMSO solution diluted with benzene, treated with anhy. HCl	black tar obtained
Fe/HCl[47] benzene	benzene solution treated with anhy. HCl	black tar obtained
3% Na(Hg)[48] methanol	added $Hg(OAc)_2$[46]	gray solid obtained

dehydration to the corresponding maleimides by the method of Searle[49] were unsuccessful.



Another strategy to synthesize N-[2-(4-imidazolyl)ethyl]maleimide is outlined below.

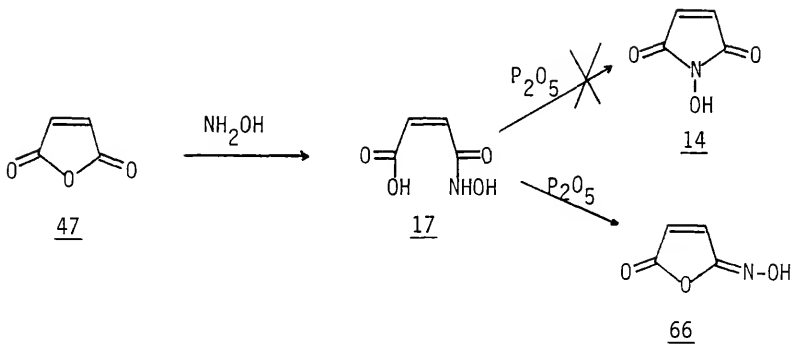


Reaction of the furan-maleic anhydride Diels-Alder adduct (8) with 20 afforded compound 24 in good yield. Dehydration of this succinamic acid derivative was also unsuccessful using the $Ac_2O/NaOAc$[49] and N,N-dicyclohexylcarbodiimide (DCC)/DMF[50] procedures.

Hydroxamic Acid --Maleimides

The simplest hydroxamic acid --maleimide is N-hydroxymaleimide (14). The acidity of hydroxamic acids is comparable to the acidity of carboxylic acids;[51] thus, one would expect hydroxamic acid groups incorporated into a polymer to be significantly ionized in neutral or basic media. As N-hydroxysuccinimide has a pka of ~6.0,[52] it was believed copolymerization of 14 would fulfill the hydroxamic acid requirement.

Ivanov et al.[53] reported the synthesis of 14, and we attempted to duplicate this synthesis as outlined below:

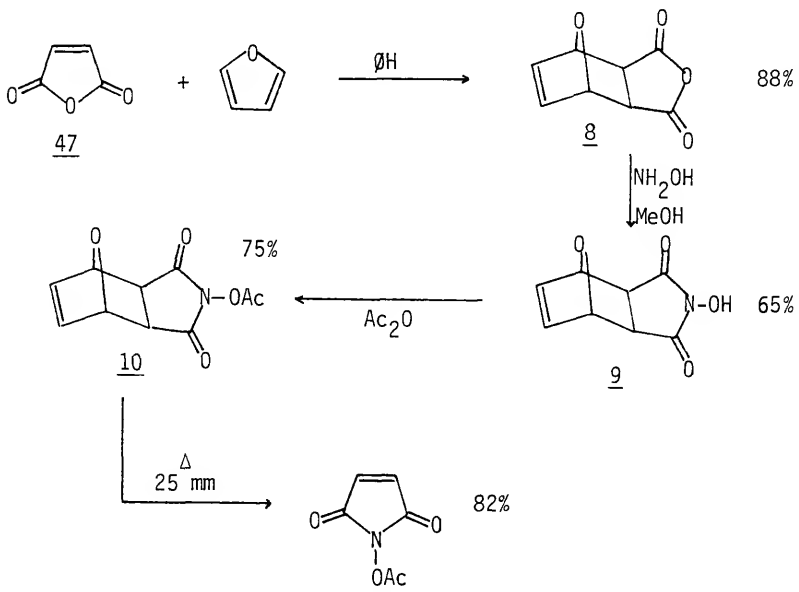


Treatment of maleic anhydride (47) with hydroxylamine afforded N-hydroxymaleamic acid (17) in 60% yield. We attempted dehydration of 17 using the $Ac_2O/NaOAc$[49] and P_2O_5/DMF[50] methods, but were unable to isolate 14. Narita et al.[50] had also studied the dehydration of 17 using the following reagents: P_2O_5, $SOCl_2$, Ac_2O, p-toluenesulfonic acid, DCC, and acetyl chloride in pyridine. Reaction of 17 with P_2O_5 in DMF gave N-hydroxyisomaleimide (66) and not 14 as reported by Ivanov et al. Neither 14 or 66 was obtained by Narita et al. using the other above mentioned dehydration reagents.

In another publication by Narita et al.[15] was described the synthesis of N-acetoxymaleimide (11). We felt this monomer would better suit our needs than 14 itself. It has been observed by Kunitake et al.[43] that polymerization of monomers containing unprotected hydroxamic acid groups is difficult, because the hydroxamic acid group is tautomeric with the nitrone structure, and nitrones are efficient free-radical trapping agents.

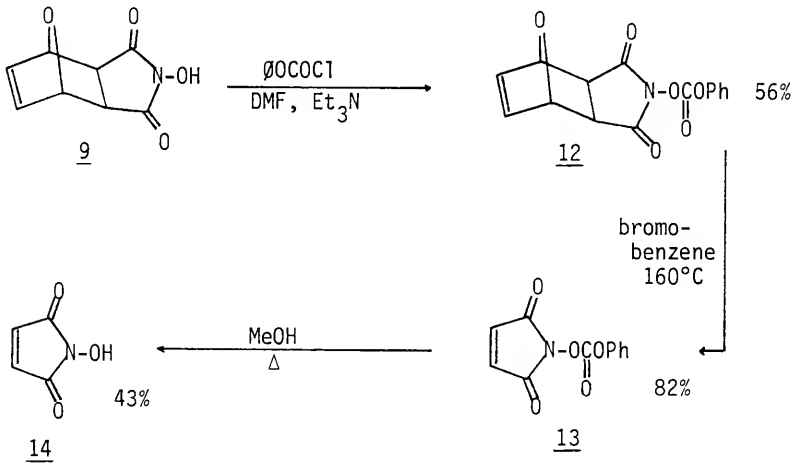
hydroxamic acid nitrone

Therefore, it seemed advisable to use a maleimide containing a protected hydroxamic acid group for polymerizations. Protected maleimide (11) was synthesized via the steps outlined below. The yields indicated are those obtained in this laboratory.



The reverse Diels-Alder reaction of N-substituted maleimide adducts of furan is a powerful method for synthesis of N-substituted maleimides which cannot be obtained from the direct dehydration of the corresponding maleamic acids. Indeed, isomaleimide formation was not a side reaction in this synthesis.

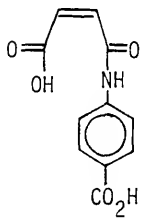
The preparation of N-hydroxymaleimide (14) itself was described in two publications by Akiyama et al.[17,54] This synthesis also employed the furan-maleic anhydride Diels-Alder adduct as a means to obtain maleimide (13), the methanolysis of which gave 14. The reaction scheme along with yields obtained in this laboratory is outlined below.



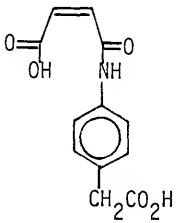
Carboxylic Acid --Maleimides

The carboxylic acid group has been incorporated into synzymes[55,56] and also plays a key role in the "charge-relay system" of the natural enzyme chymotrypsin.[57] We attempted the synthesis of carboxylic acid containing maleimide monomers for two reasons: evaluation of bifunctional synzymes containing the carboxylic acid residue, and conversion of the carboxylic acid group via its N-hydroxysuccinimide ester[58] to a hydroxamic acid group.

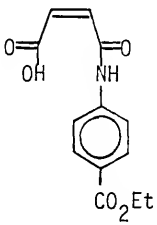
The direct dehydration of maleanilic acids 22 and 23 were unsuccessfully carried out using the following reagents: $Ac_2O/NaOAc$,[49] DCC/DMF,[50] DCC/CH_2Cl_2,[59] and refluxing xylene.[60] On the other hand, maleanilic acid 15 was cleanly dehydrated to maleimide 16 in 83% yield using $Ac_2O/NaOAc$.



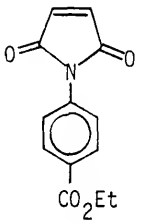
22



23

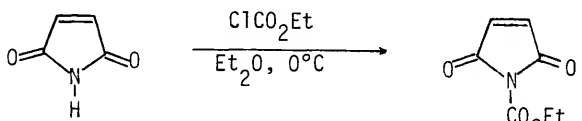


15



16

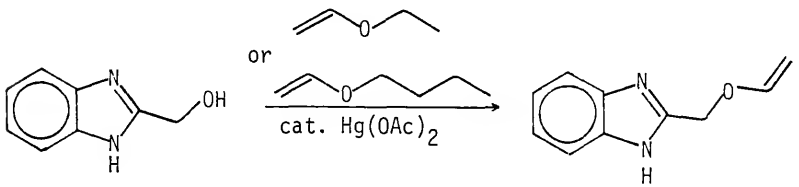
Clearly, the presence of the second carboxyl group in 22 and 23 is in some way responsible for the inability to effect dehydration. Other workers[61] have reported on the inability to dehydrate amino acid-maleamic acids, and the synthesis of maleoylamino acids by modification of maleimide itself has only recently been reported.[20,62] Using this former procedure, N-carbethoxymaleimide (18) was synthesized in 44% yield.



18

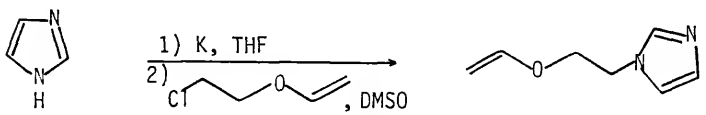
Imidazole -- Vinyl Ethers

Although imidazole substituted vinyl ethers have not been reported in the literature, we were able to synthesize these compounds in some cases via modification of standard procedures. Our first attempt involved the vinyl transetherification reaction as reported by Watanabe and Conlon.[63]



Thus, 2-benzimidazole methanol was treated with an excess of ethyl vinyl ether or n-butyl vinyl ether at reflux in the presence of a catalytic amount of mercuric acetate. ^{1}H NMR analysis of the reaction mixtures (upon removal of excess vinyl ether) indicated that no reaction had taken place. A possible explanation for the failure of the desired reaction to work was the observation that 2-benzimidazole methanol was largely insoluble in both ethyl vinyl ether and n-butyl vinyl ether.

A different approach to this class of compound was more fruitful.

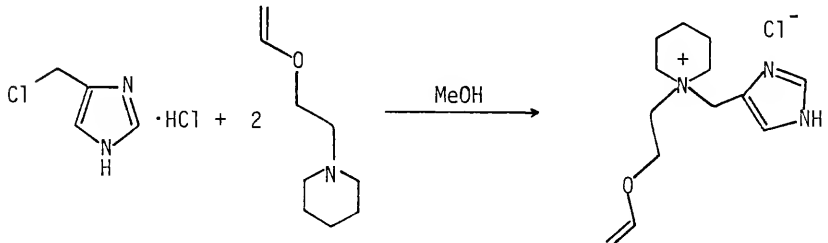


28

Alkylation of the potassium salt of imidazole with 2-chloroethyl vinyl ether (CEVE) in DMSO afforded desired N-(β-vinyloxyethyl)imidazole (28)

as a colorless oil in 46% yield. Vinyl ether (28) was also synthesized via the sodium salt (NaH) of imidazole, albeit in lower yield (25-30%). Although 28 is a tertiary (N-substituted) imidazole, it was believed that the position of substitution would not significantly hinder this compound's ability to catalyze deacylation of the acylhydroxamate intermediate in esterolysis reactions.[42]

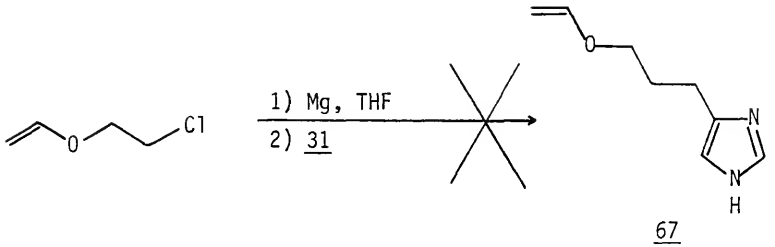
A 4(5)-substituted imidazole--vinyl ether, β-vinyloxyethyl(imidazol-4ylmethyl)piperidinium chloride (30), was synthesized, and its ability to copolymerize with maleimide (16) was studied.



31 29 30

Reaction of N-(β-vinyloxyethyl)piperidine (29) and 4-(chloromethyl)-imidazole hydrochloride (31) gave 30 in yields of 70-80%. Vinyl ether 30 proved very difficult to purify, requiring repeated treatment with Na_2CO_3 in methanol and precipitation into ethyl ether to remove excess 29 as its free base. Furthermore, 30 was isolated as a gummy hygroscopic solid, and attempts to isolate 30 as its hydrochloride salt resulted in rapid hydrolysis of the vinyl ether group. Vinyl ether 30 was consequently used for copolymerization studies in free base form. Despite prolonged drying in vacuo, a major contaminant in 30 was ethyl ether as determined from ^1H NMR.

Another attempt at preparation of a 4(5)-substituted imidazole--vinyl ether is outlined below.

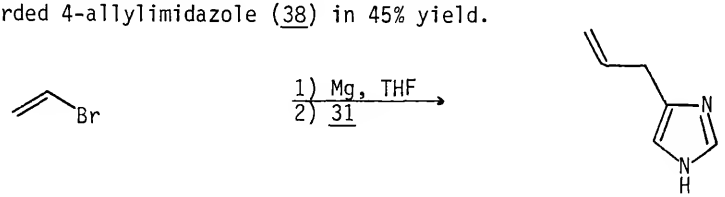


67

The covalently bound chlorine atom in 31 is extremely labile to nucleophiles as reported by Turner et al.[29] It was postulated that vinyl ether 67 could be obtained by slowly adding 31 to a solution of 2-chloroethylmagnesium chloride. The free base of 31 is not stable, presumably due to self-condensation, which necessitated the use of 31. However, CEVE reacted with Mg turnings in THF to give an insoluble Grignard reagent. Addition of 31 resulted in no 67 being formed.

Other Imidazole Monomers

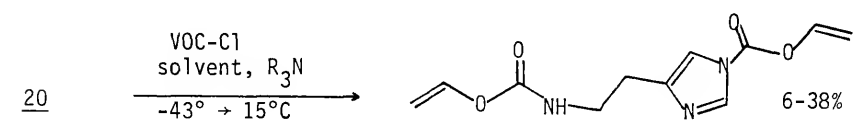
A search for other imidazole containing monomers which might alternately copolymerize with N-substituted maleimides was conducted. Convinced that the reaction of a Grignard reagent with 31 as described above should work, the reaction of vinylmagnesium bromide with 31 afforded 4-allylimidazole (38) in 45% yield.



This compound had previously been synthesized and characterized by Begg et al.[33] from the pyrolysis of N-allylimidazole, giving approximately equal amounts of 2- and 4-allylimidazole. Our synthesis thus represents a new and regiospecific method of obtaining 38. In view of the report that allylphenols and N-substituted maleimides copolymerize in alternate fashion,[11] copolymerization studies with 38 were carried out.

It was postulated that N-substituted maleimides might also alternately copolymerize with vinyloxycarbonyl-substituted imidazoles, due to similarities in structure between vinyl ethers, vinyl esters, and vinyl carbamates. An empirical comparison of monomer reactivities can be found in the Q-e scheme introduced by Alfrey and Price.[64] The e value, which is a measure of monomer polarity, is positive for electron-deficient olefins such as N-phenylmaleimide (e = +3.24) and negative for electron-rich olefins such as CEVE (e = -1.58), vinyl acetate (e = -0.88), and vinyl N,N-diethylcarbamate (e = -1.10).[65] The reaction of histamine (20) and vinylchloroformate (VOC-Cl) was therefore studied.

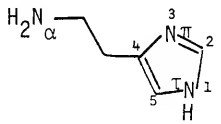
The reaction of 20 and VOC-Cl was carried out in organic solvents in the presence of an organic base over a range of temperatures.



37

In each case, the major isolable product was a diacylated histamine as evidenced by mass spectrometry (M^+ = 251), ^1H NMR (presence of 2 ABX patterns), and ^{13}C NMR (11 resonances). In the IR spectrum, carbonyl stretching frequencies were observed at 1775 and 1735 cm^{-1}.

There are three sites on 20 which might be acylated, N_α, N_π, and N_τ.[66]



Difficulty was encountered in determining the sites of substitution. Structure 37 was tentatively assigned as the N^α, N^τ isomer, although the N^α, N^π and N^α, N^α structures cannot be ruled out. The IR carbonyl stretching frequencies were especially disturbing in view of the carbonyl stretch (1710 cm^{-1}) observed for the monoacylated histamine (35). In Table III is reported the conditions and yields obtained in the reaction of 20 with VOC-Cl.

TABLE III

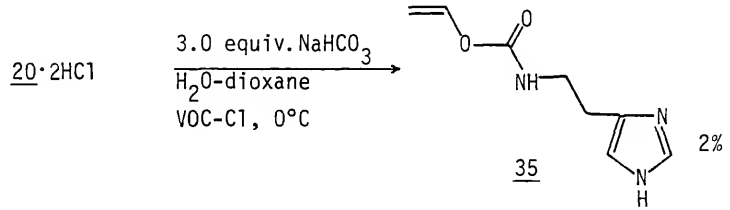
Acylation of Histamine (20) with VOC-Cl

20 (equiv)	VOC-Cl (equiv)	Solvent	Base (equiv)	T (°C)	% Yield 37[a]
1.0	1.0	dioxane	Et_3N (1.0)	12-14	6.2
1.0	0.95	$CHCl_3$[b]	Et_3N (0.95)	0-5	38
1.0	0.95	$CHCl_3$[b]	-	0	22
1.0	0.95	$CHCl_3$[b]	Et_3N (0.95)	-43	33
1.0	0.95	$CHCl_3$[b]	pyridine (XS)	-43	-

[a] yield based on VOC-Cl

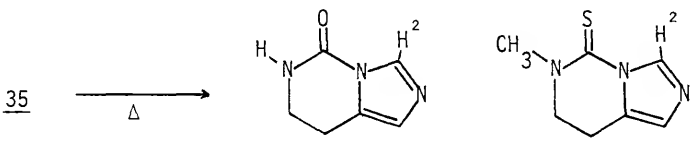
[b] ethanol-free $CHCl_3$

When the reaction of 20 and VOC-Cl was carried out under aqueous conditions, the monoacylated histamine 35 was obtained in low yield.



35

Evidence for structure assignment is based on mass spectrometry (M^+ = 181), ^1H NMR (broad one proton signals at 5.67 and 9.19 ppm), and ^{13}C NMR (8 resonances). When 35 was heated above its melting point, a reaction involving displacement of a vinyloxy group took place.

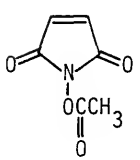


36 zapotidine

This compound was assigned structure 36 on the basis of mass spectrometry (M^+ = 137), ^1H NMR (loss of vinyl signals), and ^{13}C NMR (6 resonances). Furthermore, H-2 (the proton flanked by both nitrogen atoms of the imidazole ring) appears further downfield (8.04 ppm) than usual. Comparison with the ^1H NMR spectrum of zapotidine (H-2 = 8.42 ppm) confirms that the downfield resonance of H-2 can be attributed to the deshielding effect of the carbonyl group.[67] No copolymerization studies were conducted with 35 due to difficulties in obtaining sufficient quantities to work with. It was also suspected that 35 rearranged to 36 while being chromatographed on silica gel.

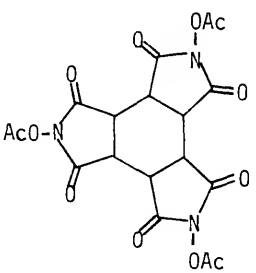
Copolymerization of Maleimides with N-(β-Vinyloxyethyl)imidazole (28)

Addition of 28 to a CH_2Cl_2 solution of 11 resulted in the immediate appearance of a blood-red color which intensified with time. No color change occurred when CEVE was added to a solution of 11. The red color was also apparent in solutions of N-vinylimidazole (46)--11 and N-methylimidazole -- 11, suggesting that the imidazole group was responsible for the red coloration. After appreciable reaction times, the red solutions were precipitated into hexanes or ether-giving pale-red solids in each case. Evaporation of the filtrate in vacuo gave an oil whose ^1H NMR spectrum revealed a preponderance of N-substituted imidazole over 11. The red solids appeared to be the same substance as judged from their IR spectra (1820, 1790, 1740, 1225, 1160 cm^{-1}) and appeared to be the homopolymer of 11. The belief that N-substituted imidazoles were catalyzing homopolymerization of 11 was demonstrated by carrying out the reaction using 0.5 mol% of 28.



11

0.005 equiv. 28

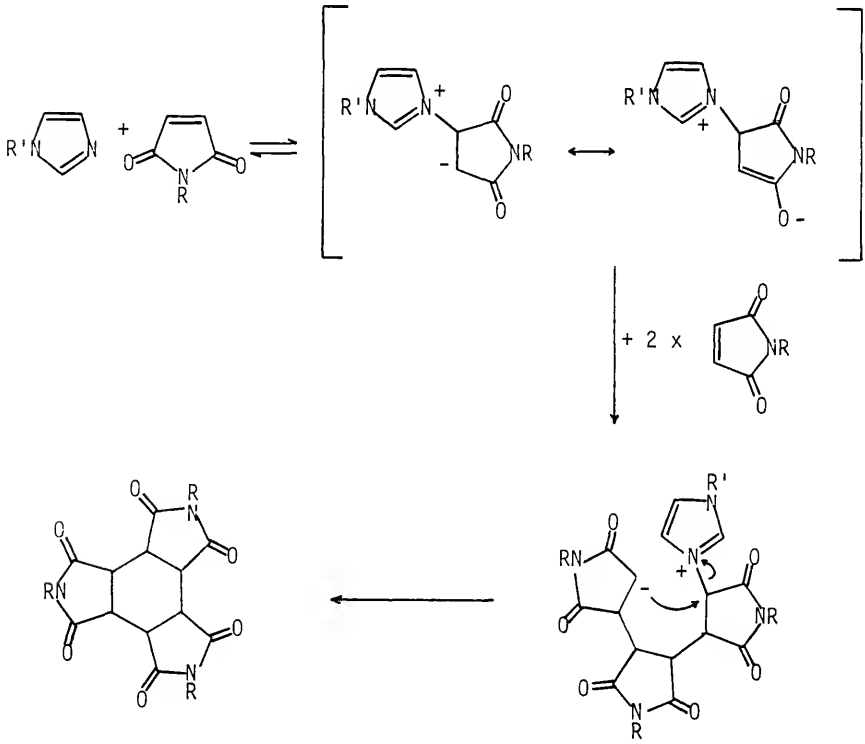


65

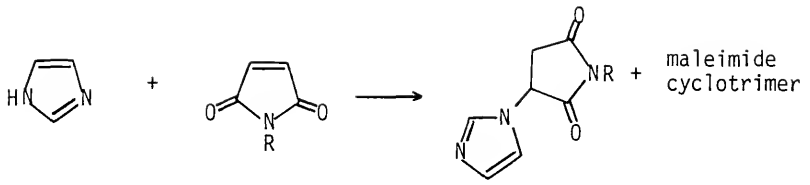
16%

Further insight as to the structure of 65 was gained by determining the molecular weight by VPO ($\overline{M}_n$ = 488). This information revealed that 65 was most likely a trimer of 11.

A search of the literature revealed that this reaction had been reported previously, and the major product was a maleimide cyclotrimer.[68] Wagner-Jauregg and Ahmed[69] invoked a zwitterionic mechanism to account for the observed product.

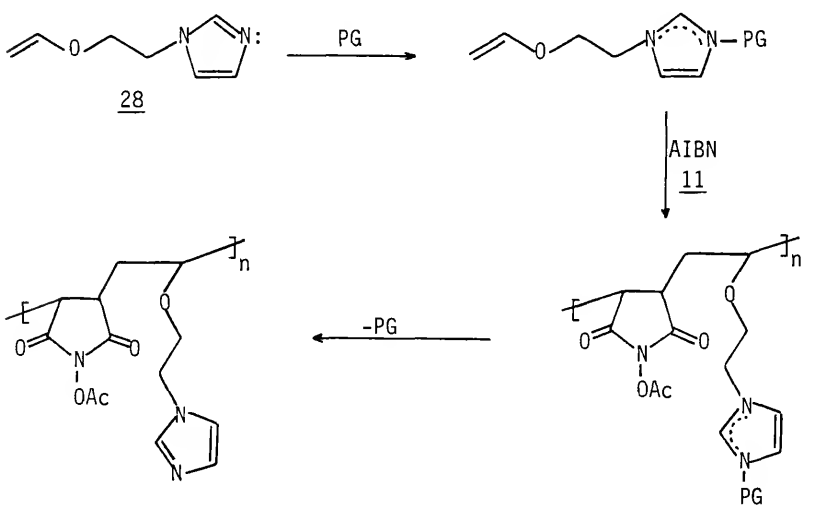


Furthermore, in addition to the desired Michael adduct, 1-25% of maleimide cyclotrimer was formed when imidazole was reacted with N-substituted maleimides in stoichiometric amounts.[68]

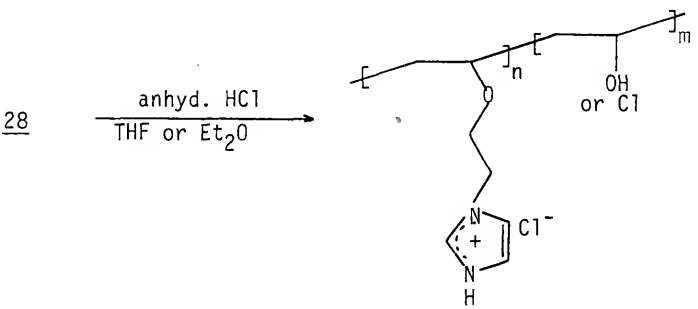


Although this information was disconcerting as far as our copolymerization strategy was concerned, we nevertheless attempted the copolymerization of 11 and 28 using AIBN as initiator. Not surprizingly, only homopolymers of 11 were obtained. Direct copolymerization of maleimides 13 and 14 with 28 were also unsuccessful.

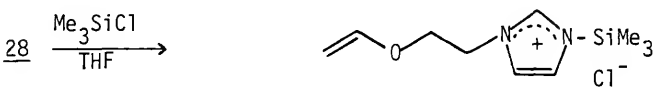
Recognizing that the cyclotrimerization reaction was responsible for the inability to obtain alternating copolymer, ways were sought to circumvent this side reaction. It was reasoned that reaction of 28 with a Lewis acid would effectively retard the imidazole residue's ability to catalyze cyclotrimerization.



The ideal protecting group (PG) would coordinate strongly enough with the imidazole moiety to preclude cyclotrimerization yet be easily removed following copolymerization. The hydrochloride salt of 28 might afford adequate protection, however when 28 was treated with anhydrous HCl in ether or THF solution, oligomerization of 28 occurred with concomitant cleavage of the vinyl ether.

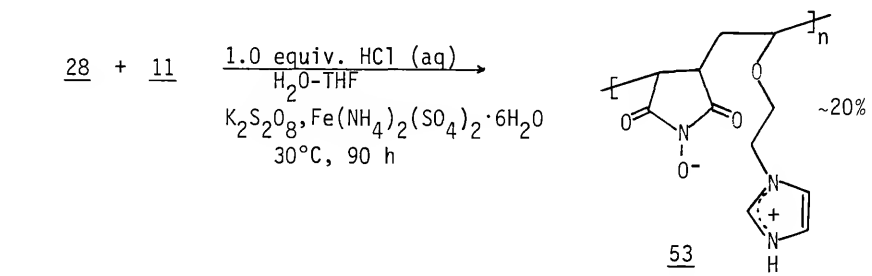


Reaction of 28 with chlorotrimethylsilane (Me_3SiCl) in THF might also be expected to provide protection.



In these experiments, either 28 or 46 was allowed to react with 1.0 equivalent of Me_3SiCl in THF in a polymerization tube under N_2. A solution of 11 or 13 and AIBN in THF was subsequently added. The appearance of a pink color probably indicated that the N-Si bond was too labile to afford adequate protection. Homopolymers of 11 and 13 were obtained in low yields.

Vinyl ether 28 and maleimide 11 were successfully copolymerized when 28 was pretreated with 1.0 equivalent of aqueous HCl. Maleimide 11 was added in acetone or THF solution to give a homogeneous mixture. Polymerization was initiated via redox conditions, $K_2S_2O_8$ and an Fe(II) salt.



53

The copolymer was purified by precipitating the reaction mixture into acetone (to remove maleimide homopolymer), redissolving the oily precipitate in aqueous HCl and dialyzing this solution against deionized water. Copolymer 53 precipitated from solution during dialysis. The IR spectrum of 53 indicated the presence of both monomers, carbonyl stretching frequencies at 1780 and 1705 cm^{-1} (indicating that the N-acetoxy group had been hydrolyzed) and C-O-C ether stretch at 1105 cm^{-1}. The carbonyl absorbances for 53 are identical to those reported for N-hydroxymaleimide -- styrene copolymer.[70] Evidence for alternation can be found in ^{13}C NMR spectrum of 53 (Figure 1). In the carbonyl region appear 3 peaks in area ratios of ~2:1:1. This is consistent with an alternating copolymer with a homogeneous sequence distribution whose stereochemistry at the succinimide unit is exclusively cis or trans and whose carbonyls can "see" relative stereochemistry two bonds distant. Carbon 10 can be assigned as the upfield doublet

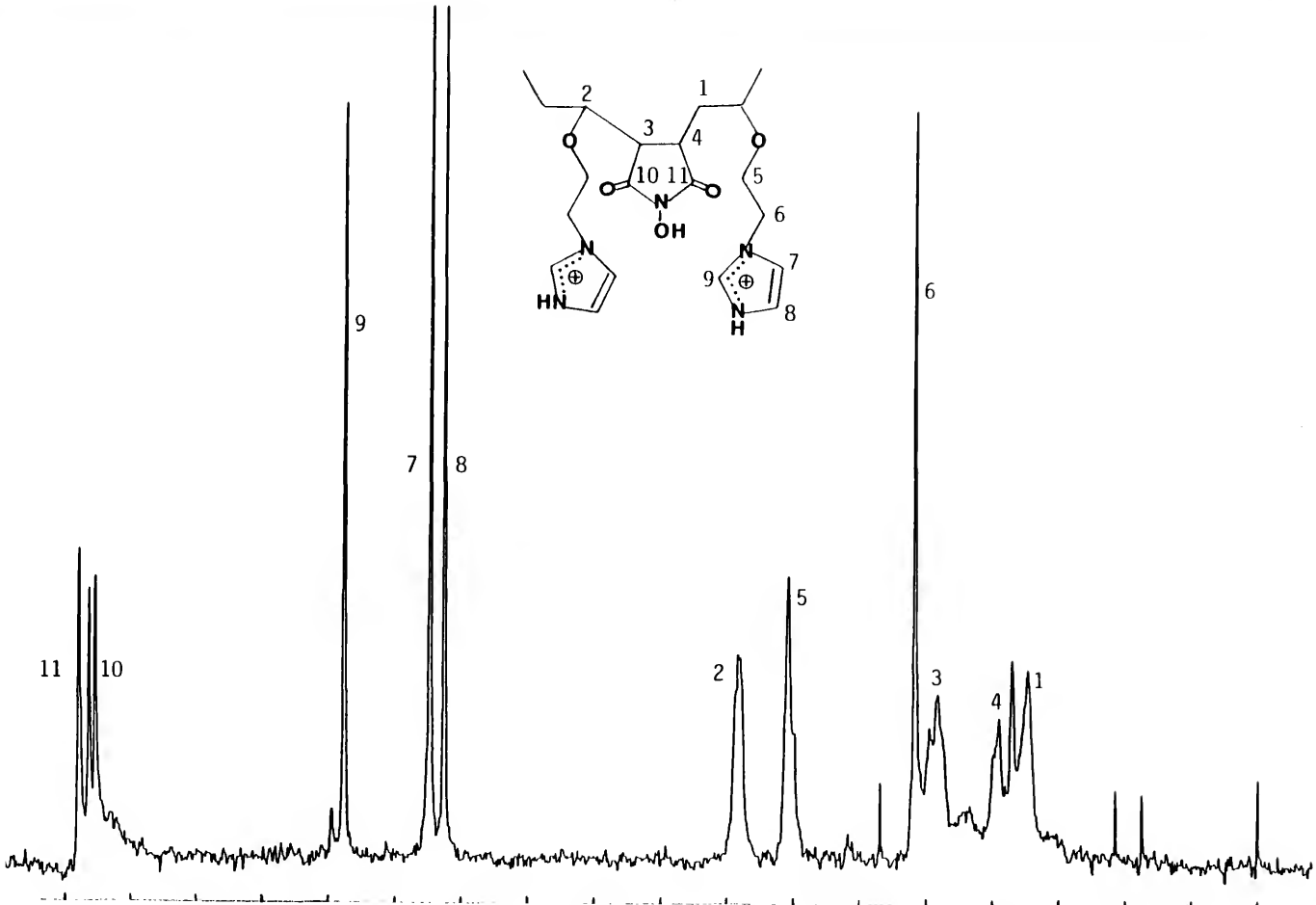


Figure 1. Proton decoupled ^{13}C NMR spectrum of copolymer 53 in D_2O-HCl, 70°C.

reflecting random relative stereochemistry between C-2 and C-3. Carbon 11 then appears as a singlet as a result of its inability to "see" relative stereochemistry three bonds distant. The carbonyl region of 53 is analogous to the carbonyl region of N-phenylmaleimide --2-chloro ethyl vinyl ether alternating copolymer as reported by Olson.[12] Indeed, a more complete explanation of this reasoning can be found in this work.[12]

The assignment given carbonyl carbons 10 and 11 can also be rationalized by empirical chemical shift parameters first reported by Grant and Paul.[71] Substituents other than protons which are situated α or β to a carbon of interest cause a downfield shift (~9 ppm) relative to a similar compound without substitution. Substituents other than protons which are situated γ to a carbon of interest cause an upfield shift (~2 ppm). Examination of the copolymer structure reveals that carbons 10 and 11 have equal numbers of α and β substituents. Carbonyl 10, however, has an addition γ substituent by virtue of branching at C-2. Therefore, C-10 should appear further upfield than C-11.

The resonances between 120 and 140 ppm were assigned to the carbons of the imidazole ring. The signal appearing furthest downfield was assigned to C-9, while no distinction could be made between C-7 and C-8.

The resonances between 70 and 80 ppm are typical for carbon atoms alpha to an oxygen atom. Differentiation between C-2 and C-5 could be made after examination of an off-resonance spectrum (Figure 2). The signal furthest upfield appeared as a triplet (C-5) and the downfield

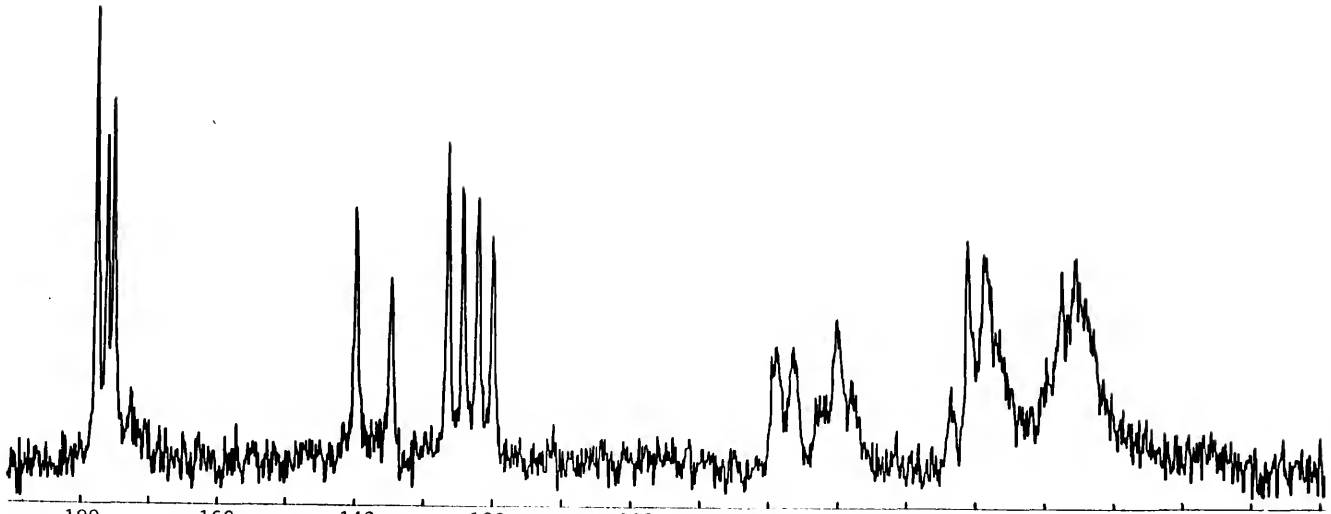
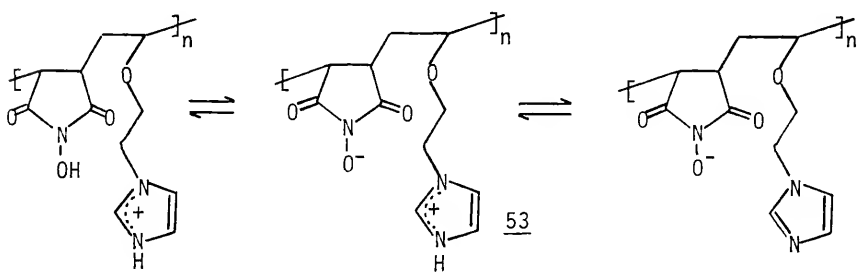


Figure 2. Off-resonance decoupled ^{13}C NMR spectrum of copolymer 53 in D_2O-HCl, 60°C.

signal as a doublet (C-2). Carbon-6 also appeared as a triplet in the off-resonance ^{13}C spectrum.

Further evidence for alternation can be obtained from the chemical shifts of the succinimide backbone carbons, C-3 and C-4. In the ^{13}C NMR spectrum of N-hydroxymaleimide homopolymer (50), the backbone carbons appear as a broad singlet centered at ~42 ppm. In copolymer 53 the succinimide backbone carbons appear as two signals at ~39 and ~48 ppm. Carbon-4 was assigned to the upfield signal on the basis of having one additional γ substituent vs. C-3. In addition, C-3 has one additional β substituent than does C-4. The broad hump appearing between C-3 and C-4 at ~42 ppm is probably attributable to the methine carbons of homomaleimide sequences. The methylene backbone carbon atom (C-1) was assigned to the signal appearing furthest upfield.

Copolymer 53 possesses very interesting solubility characteristics. As isolated, 53 is virtually insoluble in all common organic solvents, although it will dissolve in DMSO at elevated temperatures. Interestingly, 53 is water soluble at pH <3.6 and pH >7.1 but insoluble between these limits. This solution behavior led us to believe that 53 is a polyampholyte.



53

Solubility is attained when the copolymer is protonated or deprotonated giving rise to net positive or negative charges. Under these conditions, 53 might be expected to behave as a polyelectrolyte. At the isoelectric point, however, the attraction between oppositely charged side chains should result in tight coiling, hence a lack of solubility. In this case, an increase in the ionic strength of the medium should lead to expansion of the chains and impart water solubility. Such behavior has been noted for poly(vinyl imidazolium sulphobetaine) (PVISB) by Salamone et al.[72]

The solubility of 53 in various salt solutions is shown in Table IV. It can be seen that certain salts are more effective than others with respect to dissolving power. Interestingly, both sat. LiCl and sat. LiBr completely dissolve 53, while 5.0 M LiCl only partially dissolves 53. It is also not clear why 53 is soluble in sat. NaI, partially soluble in sat. NaCl, and apparently insoluble in sat. NaBr.

TABLE IV

Solubility of 53 in Salt Solutions

Cation \ Anion	Cl^-	Br^-	I^-	BF_4^-
Li^+	sat. + 5.0M P	sat. +		
Na^+	sat. P	sat. -	sat. + 3.0M P	
K^+		sat. P	sat. -	sat. -

sat. = saturated solution
+ = soluble
- = insoluble
P = partially soluble

Salamone et al.[72] found that large cations, e.g. K^+, and large anions, e.g. ClO_4^-, were more effective solubilizing ions than were smaller ions. Minimum salt concentrations were of the order of 0.03-0.52M for PVISB, while saturated salt solutions were necessary to impart solubility to 53.

Determination of the molecular weight of 53 proved difficult, and the results were ambiguous. VPO analysis in DMSO at 100°C gave $\overline{M}_n \cong$ 500 g/mol. It is believed that this number represents a minimum value as 53 was observed to decompose under similar conditions in the NMR probe. As VPO is a colligative technique, the presence of decomposition fragments would result in a lower $\overline{M}_n$ than expected. A maximum molecular weight value was calculated from end group analysis (elemental analysis for S). Presumably the initiating species in the redox system employed is the sulfate radical anion.[73]

$$S_2O_8^{2-} + Fe^{+2} \longrightarrow SO_4^{2-} + SO_4^{\cdot-} + Fe^{+3}$$

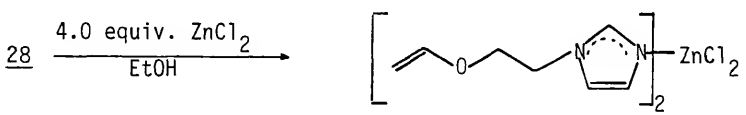
Assuming the incorporation of one sulfate group per polymer chain, from calculation of an empirical formula a molecular weight of ≅ 6000 g/mol was derived.

The intrinsic viscosity [η] of 53 was determined in 0.1N HCl at 30.0°C to be 0.112 dL/g. Although this value cannot be directly related to molecular weight, it is an indication of the hydrodynamic volume of 53. Of course the size of the polymer chains should vary with the pH of the medium, and one would expect a change in [η] dependent on the degree of ionization of imidazole groups.

Gel permeation chromatography of 53 revealed two components in a 3:1 area ratio, the larger component having the shorter retention volume. The presence of two components precluded accurate molecular weight determination, as the individual viscosities could not be determined independently. It was suspected that the minor component was attributable to N-hydroxymaleimide homopolymer (50), which we were unable to separate from 53.

In another set of experiments, 28 was mixed with $ZnCl_2$ in THF before addition of AIBN and 11. In this case, a white-THF insoluble powder was obtained after only a few hours at 60°C. The IR spectrum of this material indicated that both 11 and 28 had been incorporated as evidenced by carbonyl stretching frequencies at 1818, 1785 and 1730 cm^{-1} and C-O-C ether stretch at 1110 and 1095 cm^{-1}. The incorporation of $ZnCl_2$ into this material was inferred by elemental analysis for Cl (9.22%). Elemental analysis was reasonably consistent with a structure containing two equivalents of 28 and two equivalents of 11 per equivalent of $ZnCl_2$.

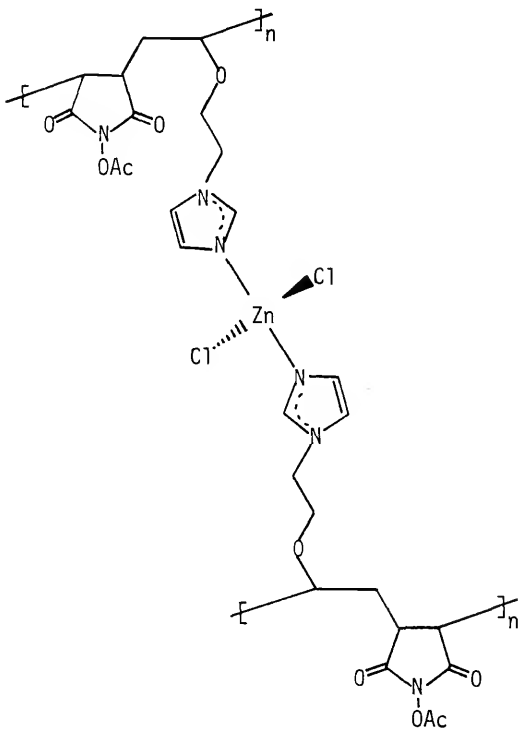
Reaction of 28 with four equivalents of $ZnCl_2$ in ethanol gave, after dilution with ether, an 89% yield of 33, mp 78.5-80°C.



33

The stoichiometry of this complex was determined from elemental analysis and is consistent with N-alkylimidazole -- $ZnCl_2$ complexes studied by Welleman et al.[32] On the basis of the structure of 33 and the

elemental analysis data for the $ZnCl_2$ copolymer, the latter's repeating unit was assigned structure (54).



54

This crosslinked structure accounts for the insolubility of 54 in common organic solvents. However, 54 dissolves in warm DMSO (~60°C), presumably giving a DMSO-$ZnCl_2$ [74] complex and free copolymer. The [η] of a DMSO solution of 54 at 30.0°C was equal to 0.043 dL/g. Copolymer 54 is also soluble in water at pH >13 (NaOH) and pH <3.5 (HCl).

The proton decoupled ^{13}C NMR spectrum of 54 is shown in Figure 3. Similarities to the ^{13}C NMR spectrum of 53 (Figure 1) are apparent, and the assignment of carbon atoms is the same as in 53. Two additional peaks - one in the carbonyl region and one at ~22 ppm - can probably be assigned to acetic acid (from hydrolysis of the N-acetoxy group). The resonance at ~42 ppm most likely indicates the presence of homomaleimide sequences.

Copolymerization of Fumaronitrile (45) and Diethylfumarate (44) With N-(β-Vinyloxyethyl)imidazole (28)

In view of the fact that 28 would not cleanly copolymerize with 11 or 13, it was decided to attempt copolymerization of 28 with fumaronitrile (45) and diethylfumarate (44). As both latter monomers are electron deficient, 45 (e = +2.73)[65] and 44 (e = +2.26),[65] it was believed copolymerization with vinyl ether (28) might afford alternating copolymers. Indeed, alternating copolymers have been obtained from copolymerization of both 45 and 44 with N-vinylcarbazole[75] (e = -1.29).[65] It was envisioned that hydroxamic acid groups could be introduced following copolymerization via transformation of the nitrile and ester groups.

Copolymerizations of 28 with 44 and 45 were carried out in solution at 60°C using AIBN as initiator. Low yields of low molecular weight copolymers were obtained in each case. The ^{1}H NMR spectrum of 56 indicated that the copolymer was rich in diethylfumarate (~2.3:1) as determined by comparing the integration of methyl protons to imidazole protons. These data are supported by elemental analysis for nitrogen, the value obtained being ~2.8% lower than expected for a 1:1

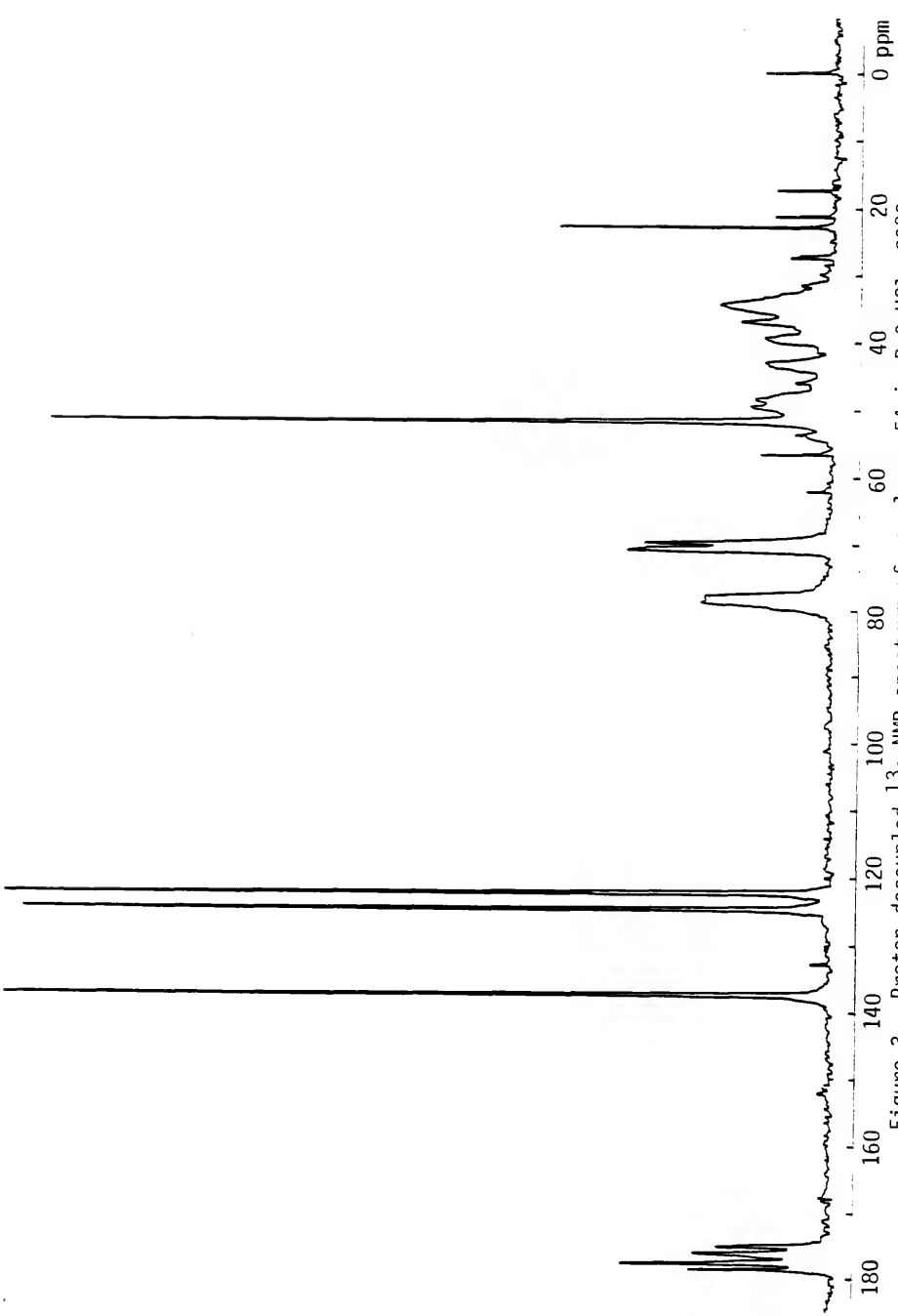
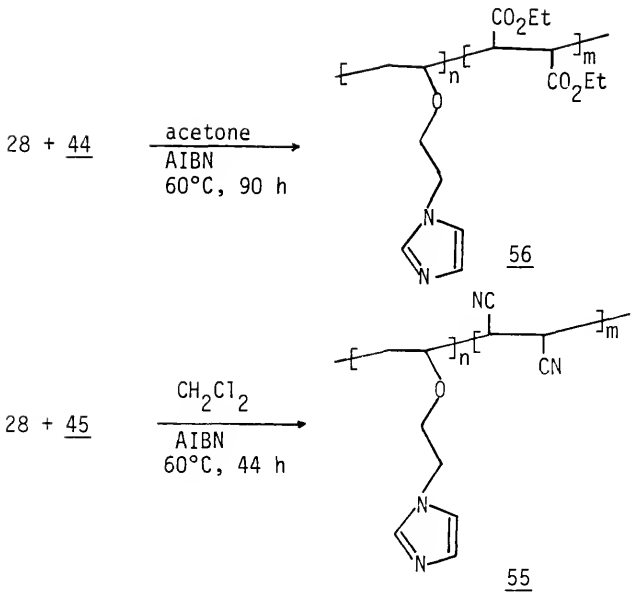


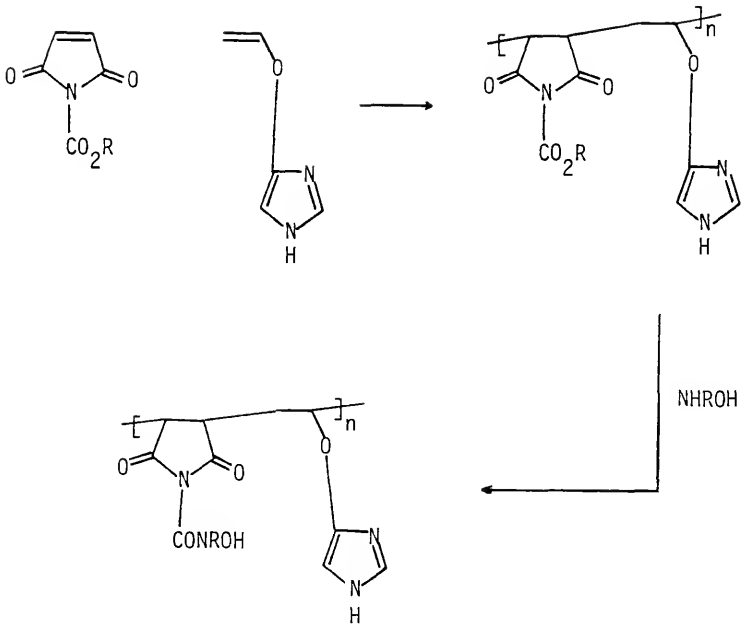
Figure 3. Proton decoupled ^{13}C NMR spectrum of copolymer 54 in D_2O-HCl, 80°C.



copolymer. Copolymer 55 was suspected of being rich in fumaronitrile on the basis of nitrogen analysis, found to be ~2.7% higher than expected for a 1:1 alternating copolymer. Since alternating copolymers were not obtained in either case, no attempt was made to transform either the nitrile or ester group to a hydroxamic acid group.

Copolymerization of Maleimide (16) With β-Vinyloxyethyl(imidazol-4ylmethyl)piperidinium Chloride (30)

After it had been established that copolymer (53) was not an efficient catalyst, new catalysts were sought. It was proposed that a better catalyst could be obtained by changing the nature of the hydroxamic acid group and by varying the position of substitution on the imidazole ring. Our strategy involved copolymerizing a 4(5)-imidazole-substituted vinyl ether with an ester-maleimide and converting the ester group to a hydroxamic acid group following the copolymerization step.



To this end, maleimide (16) and vinyl ether (30) were synthesized and their copolymerization attempted. Because of the extreme rapidity with which 30 hydrolyzed under acidic conditions, no copolymer was formed under the redox conditions under which 11 and 28 were successfully copolymerized. The hydrophobic nature of 16 necessitated the use of a water miscible organic solvent to obtain a homogeneous reaction mixture. Addition of an organic solvent to an acidic-aqueous solution of 30 resulted in an immediate exothermic reaction probably indicative of hydrolysis of the vinyl ether group. Therefore, it was believed that complete hydrolysis took place before any copolymerization could occur.

When 16 and 30 were allowed to react in organic solvents using AIBN as initiator (no acid present), only homopolymers of 16 were obtained. In Table V is summarized our attempts to copolymerize 16 and 30.

TABLE V

Copolymerization of β-Vinyloxyethyl(imidazol-4ylmethyl)piperidinium Chloride (30)

Comonomer (mol ratio)	Initiator (mol ratio)	Reaction Conditions[a]	Yield and Comments
16 (0.80)	$K_2S_2O_8/Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ (0.016)	1.0 equiv. HCl EtOH/H_2O 33°C, 68 h	- no copolymer obtained - vinyl ether cleaved
16 (0.60)	$K_2S_2O_8/Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ (0.015)	1.0 equiv. HOAc(aq) DMSO 32°C, 45 h	- no copolymer obtained - recovered unreacted 16 - vinyl ether cleaved
16 (0.50)	AIBN (0.015)	CH_3CN 60°C, 42 h	- low yield of homopolymer (51)
16 (0.60)	AIBN (0.012)	DMF 100°C, 24 h	- high yield of homopolymer (51)

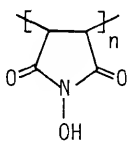
[a] carried out in glass tubes - solutions degassed and sealed under vacuum

Copolymerization of Maleimide (11) with 4-Allylimidazole (38)

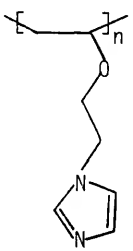
Copolymerization of 38 and 11 was attempted with and without acid protection of 38. In only one case was a copolymer obtained (as evidenced by the presence of imidazole and carbonyl resonances in the ^{13}C NMR spectrum) in low yield. It could not be determined whether the copolymer was random or alternating in structure. In any case, the IR spectrum indicated that the N-acetoxy group had been hydrolyzed to the N-hydroxy group (1780, 1710 cm^{-1}). Reaction conditions and a summary of results are given in Table VI.

Homopolymers

In order to more fully understand the catalytic activity exhibited by copolymer 53, we desired the homopolymers of 14 and 28 for comparison purposes.



50



52

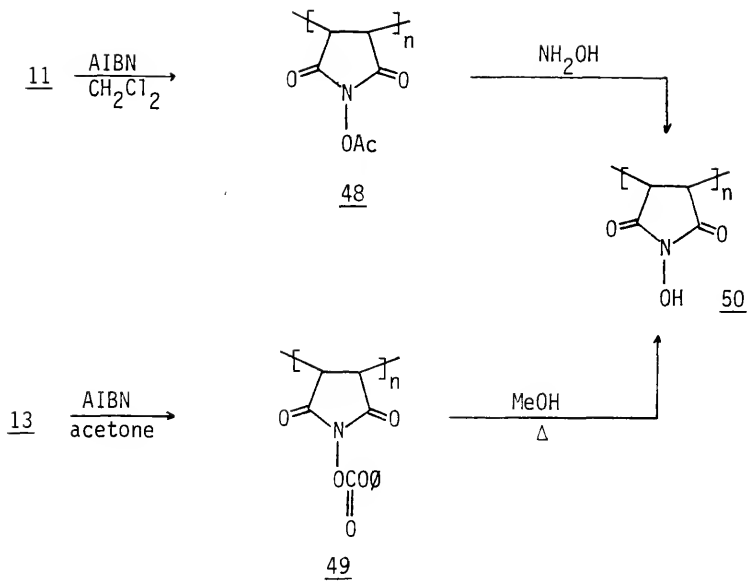
Poly(N-hydroxymaleimide) (50) was synthesized via two routes as shown below. Polymerization of 11 with AIBN in CH_2Cl_2 afforded homopolymer (48) in 80% conversion, whose molecular weight was determined by VPO ($\overline{M}_n$ = 3850 g/mol). Treatment of 48 with hydroxylamine gave 50. Homopolymer (50) exhibited carbonyl stretching frequencies of 1785 and 1770 cm^{-1} and broad resonances in the ^{13}C NMR spectrum centered at 42.0 and 172.7 ppm.

TABLE VI

Copolymerization of 4-Allylimidazole (38)

Comonomer (mol ratio)	Initiator (mol ratio)	Reaction Conditions[a]	Yield and Comments
11 (1.0)	$K_2S_2O_8/Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ (0.02)	1.0 equiv. HCl H_2O/acetone 50°C, 90 h	- no copolymer obtained
11 (0.75)	$K_2S_2O_8/Fe(NH_4)_2(SO_4)_2 \cdot 6H_2O$ (0.01)	1.0 equiv. HCl H_2O/acetone 30°C, 72 h	- <10% of copolymer (degree of alternation unknown) - acetoxy group hydrolyzed
11 (1.0)	t-BuOOH (0.01)	H_2O/acetone 60°C, 93 h	- no copolymer obtained
11 (1.0)	AIBN (0.01)	CH_2Cl_2 70°C, 19 h	- no copolymer obtained
11 (1.0)	AIBN (0.03)	1.0 equiv. HCl EtOH 60°C, 68 h	- low yield of N-hydroxymaleimide homopolymer (50)

[a] carried out in glass tubes - solutions degassed and sealed under vacuum



Homopolymer (50) was more conveniently prepared from the methanolysis of 49 in 88% yield. Polymerization of 13 with AIBN in acetone afforded 49 in 87% conversion. The proton decoupled ^{13}C NMR spectrum of 50 is shown in Figure 4.

Homopolymerization of 28 was not as straightforward. Although the polymerization of vinyl ethers with cationic initiators is well documented,[76-78] we were unable to obtain 52 in high conversion or in moderate molecular weight. The low yields obtained under cationic conditions might be due to the imidazole's ability to coordinate with the initiator, thereby rendering the latter ineffective.

Not surprisingly, 52 was not obtained by use of free-radical initiators. Table VII contains a summary of initiators, reaction conditions and results.

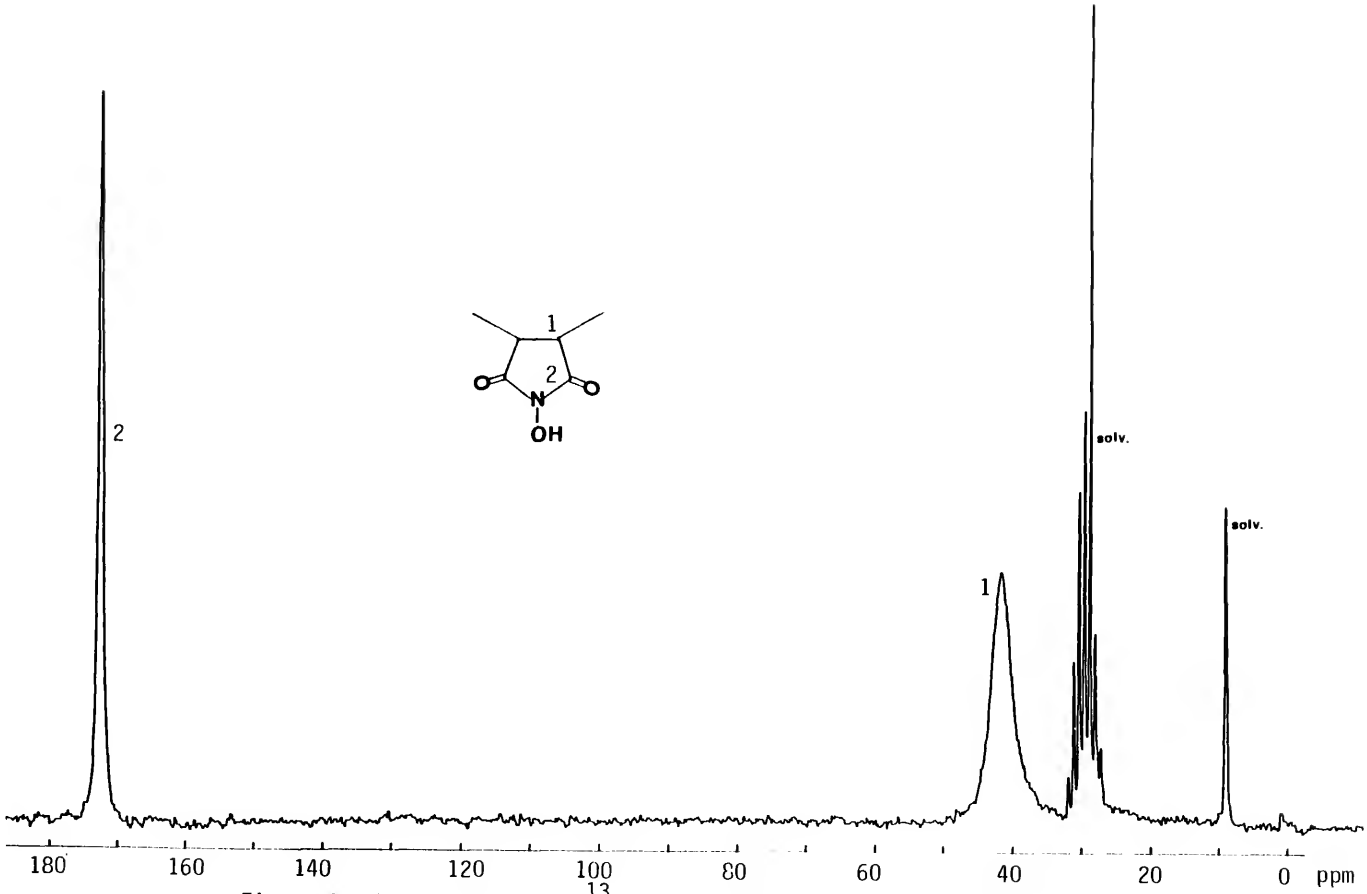


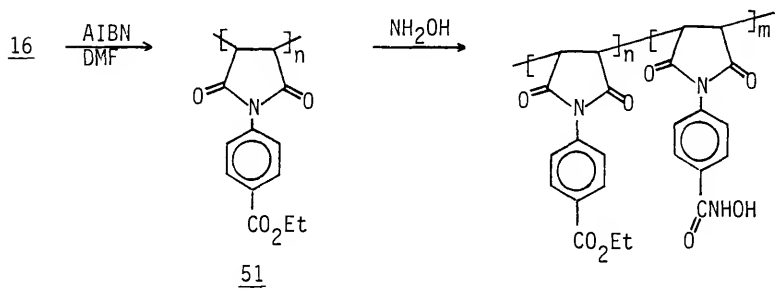
Figure 4. Proton decoupled ^{13}C NMR spectrum of copolymer 50 in acetone-d_6.

TABLE VII

Homopolymerization of N-(β-Vinyloxyethyl)imidazole (28)

Initiator (mol ratio)	Reaction Conditions	Yield and Comments
BF_3(g) (2 mL)	toluene, -78°C, N_2	- low yield of oligomers - rapid termination
$BF_3 \cdot Et_2O$ (~0.05 mL)	pentane-benzene, -78°C initiator introduced on solid CO_2,[79] 1 h	- low yield of oligomers
$Ph_3C^+ SbCl_6^-$ (0.01)	CH_2Cl_2, degassed, -42°C → -20°C	- ~1%, C-O-C stretch at 1090 cm^{-1}
I_2 (0.07)	bulk, -10°C,[80] 45 days	- obtained solid, 54% incorporation of I
I_2 (0.003)	$CHCl_3$, 0°C, N_2,[81] 20 h	- no polymer
AIBN (0.01)	acetone, 60°C, degassed, 800 h	- no polymer
$K_2S_2O_8$ (0.01)	1 equiv. HCl (aq), THF, 35°C, 68 h	- no polymer

N-(4-Carbethoxyphenyl)maleimide (16) was homopolymerized with AIBN in DMF to give 51 in 61% conversion. Surprisingly, insoluble polymers resulted when the polymerization was carried out with AIBN in CH_2Cl_2 or acetone. Reaction of 51 with hydroxylamine resulted in only partial conversion of ester to hydroxamic acid groups as judged by elemental analysis for nitrogen.



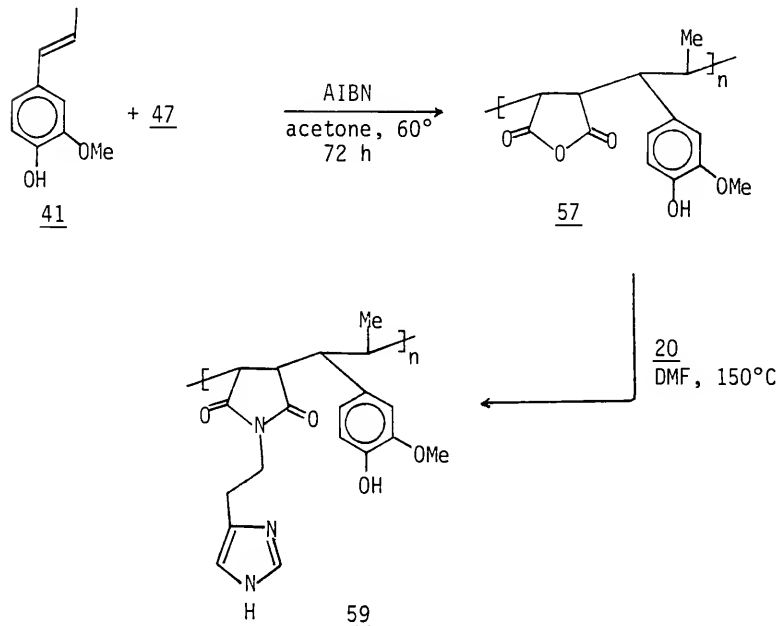
51

Copolymerization of Propenylphenols With Maleic Anhydride (47) and N-Ethylmaleimide (43)

In light of the study by Overberger et al.[7] (p. 6), which demonstrated cooperative interactions between imidazole and phenol groups, it was decided to synthesize and evaluate alternating copolymers containing these functional groups. As considerable difficulty was encountered with the direct polymerization of imidazole containing monomers, the imidazole group was introduced into pre-existing alternating copolymers.

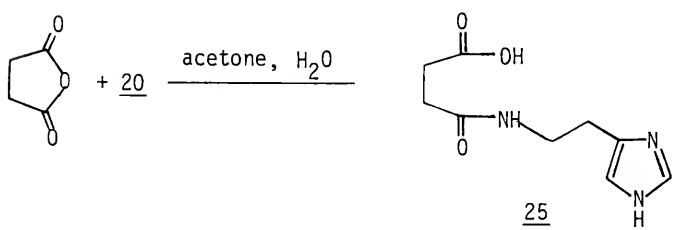
It was reported by Iwabuchi et al.[82] that a 1:1 alternating copolymer resulted from copolymerization of isoeugenol (41) and maleic

anhydride (47). This reaction was repeated in this laboratory and gave copolymer 57 in 62% conversion. Treatment of 57 with histamine (20) in refluxing DMF gave a quantitative yield of copolymer 59 containing both phenol and imidazole functionalities.

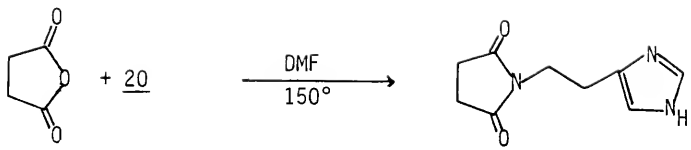


41 57 59

In order to show that 59 contained succinimide and not succinamic acid units, model compounds 25 and 26 were synthesized.



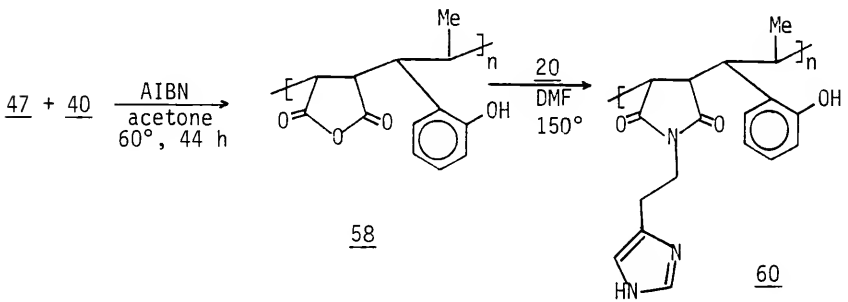
25



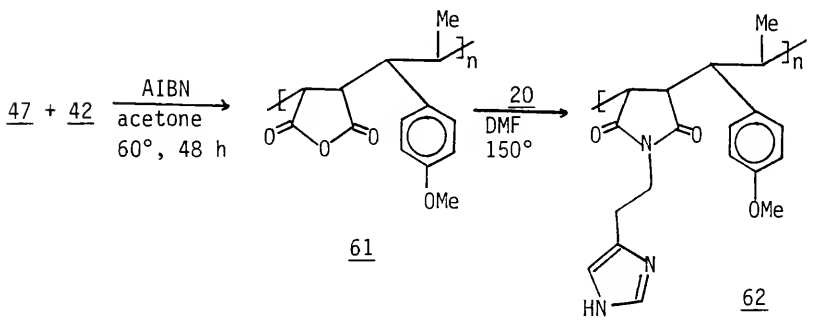
26

Imide (26) displayed carbonyl stretching frequencies at 1765 and 1690 cm^{-1} which were in agreement with those of copolymer 59. Compound 25 displayed IR absorptions at 1635 and 1610 cm^{-1}.

Copolymers 58 and 60 were synthesized in a similar manner, as were 61 and 62.

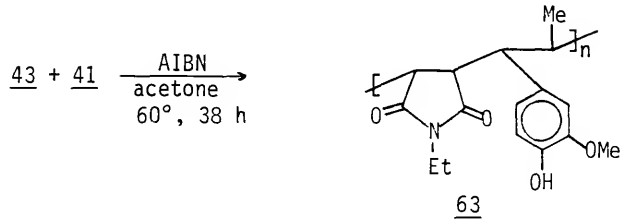


58 60

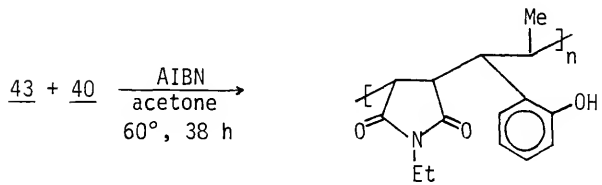


61 62

It was desired that either 59 or 60 would exhibit cooperativity in the hydrolysis of an ester substrate, while 62 would exhibit catalysis by the imidazole group only. Copolymers 63 and 64 were synthesized to demonstrate catalysis by the phenol group only.



63



64

Although we were unable to prove spectroscopically the alternating nature of these copolymers, other observations point toward a nearly alternating structure. Mixing 47 or 43 with 40, 41, and 42 produces a yellow color, probably indicative of a charge-transfer complex.[11] Iwabuchi et al.[82] have shown that 41 and 47 form a 1:1 copolymer regardless of monomer feed ratios. Elemental analysis of copolymer 61 gives excellent agreement for a 1:1 copolymer, although the others do not. Finally, neither propenylphenols nor 47 homopolymerize under the copolymerization conditions employed.

A list of some properties of copolymers 57 → 64 are presented in Table VIII.

TABLE VIII

Properties of Copolymers 57 → 64

Copolymer	$\overline{M}_n$ (g/mol)	[η] acetone (dL/g) 30.0°C	Conversion (wt%)	IR Carbonyl Stretch (cm^{-1})
57	6,950	0.183	62	1855, 1775
58	21,200	0.231	87	1855, 1770
59	-	-	100	1765, 1690
60	-	-	88	1765, 1690
61	-	-	50	1860, 1780
62	-	-	69	1770, 1695
63	25,200	0.276	88	1770, 1690
64	-	0.083	33	1770, 1695

Kinetic Studies With Imidazole, 50, and 53

Pseudo first-order kinetics were measured using imidazole, 50, and 53 as catalysts as previously described (Method A - pp. 56-59). The results obtained with these catalysts are shown in Table IX. Esterolysis in the presence of homopolymer 50 was only marginally faster than esterolysis in the absence of a catalyst. This was interpreted to mean that 50 is a poor acylating agent as compared to the phenyl hydroxamate ion studied by Kunitake et al.[43] Catalysis by 50 is slightly enhanced at higher pH, which might indicate that catalytic activity increases with the degree of deprotonation of 50. This explanation is speculative without a knowledge of the pKa of 50. In

TABLE IX

Esterolysis[a] of PNPA with Imidazole, 50, and 53

Catalyst	pH[b]	[catalyst] (N)	$k_{obs}(min^{-1})$	k_{cat} $(l \cdot mol^{-1} min^{-1})$
imidazole	7.68	7.92×10^{-4}[c]	34.9×10^{-3}	44
50	7.68	7.77×10^{-4}[d]	0.30×10^{-3}	0.39
53	7.68	7.89×10^{-4}[e]	4.41×10^{-3}	5.6
blank	7.68	-	4.92×10^{-3}[f]	-
imidazole	7.86	7.75×10^{-4}[c]	35.4×10^{-3}	45.7
50	7.86	7.73×10^{-4}[d]	0.68×10^{-3}	0.88
53	7.86	7.78×10^{-4}[e]	5.03×10^{-3}	6.5
blank	7.86	-	6.55×10^{-3}[f]	-

[a] Method A conditions: [Tris buffer] = 0.1M, μ = 0.1 (KCl), 1.5:98.5 $CH_3CN:H_2O$ (V/V), initial substrate concentration = 4.20×10^{-5}N, T = 40.0°C.

[b] pH at 25°C.

[c] Approximate concentration of imidazole groups.

[d] Approximate concentration of N-hydroxysuccinimide groups.

[e] Approximate concentration of imidazole and N-hydroxysuccinimide groups.

[f] k_{meas} of uncatalyzed reaction.

any case, the rate enhancement in the presence of 50 is small and may not be statistically important.

The catalytic activity of copolymer 53 was found to be significantly higher than 50 yet much lower than monomeric imidazole. Due to the lack of catalytic activity shown by 50, it can be concluded that neutral imidazole residues are responsible for the rate enhancement exhibited by copolymer 53. Once again, a rate enhancement was observed at higher pH, which might be related to the degree of ionization of protonated imidazole groups. Alternatively, an increase in pH might expand the polymer chains, allowing easier access of the substrate to the reaction site.

Additional kinetic studies were not carried out on this system due to the poor catalysis exhibited by 53, the knowledge that 53 was impure, the failure to obtain homopolymer 52, and the apparent lack of cooperative behavior between hydroxamic acid and imidazole groups in 53.

Kinetic Studies With Copolymers 59, 60, 62, 63 and 68 and Model Compound 26

Pseudo first-order kinetics were measured using copolymer catalysts 59, 60, 62 and 63 as previously described (Method B - pp. 56-59). The water insolubility of these copolymers necessitated the use of an 80% DMSO:H_2O (V/V) medium. Copolymer 64 was insoluble in this solvent system and therefore was not studied as a catalyst. PNPA was initially used as substrate, however its hydrolysis proceeded too slowly to obtain rate constants in a reasonable time. The more reactive substrate DNPB was subsequently used. The results obtained with these copolymer catalysts are presented in Table X and shown graphically in Figure 5.

TABLE X

Esterolysis[a] of DNPB with 26, 59, 60, 62, 63 and 68

Catalyst	pH	k_{obs} (min^{-1})	k_{cat} ($l \cdot mol^{-1} min^{-1}$)
26	8.4	4.46×10^{-3}	17.8
59	"	3.35×10^{-3}	13.4
60	"	3.62×10^{-3}	14.5
62	"	4.39×10^{-3}	17.5
63	"	0	0
68	"	4.89×10^{-3}	19.5
blank	"	1.00×10^{-3}[b]	-
26	8.9	5.30×10^{-3}	21.2
59	"	4.17×10^{-3}	16.7
60	"	4.73×10^{-3}	18.9
62	"	5.39×10^{-3}	21.5
63	"	0.06×10^{-3}	0.24
68	"	5.05×10^{-3}	20.2
blank	"	1.61×10^{-3}[b]	-
26	9.5	5.69×10^{-3}	22.8
59	"	4.93×10^{-3}	19.7
60	"	5.11×10^{-3}	20.4
62	"	5.54×10^{-3}	22.2
63	"	0.01×10^{-3}	0.04
68	"	6.78×10^{-3}	27.1
blank	"	3.84×10^{-3}[b]	-

Table X Continued

Catalyst	pH	k_{obs} (min^{-1})	k_{cat} ($l \cdot mol^{-1} min^{-1}$)
26	10.0	5.35×10^{-3}	21.4
59	"	4.21×10^{-3}	16.8
60	"	4.80×10^{-3}	19.2
62	"	5.02×10^{-3}	20.1
63	"	0.36×10^{-3}	1.44
68	"	6.91×10^{-3}	27.6
blank	"	5.79×10^{-3} [b]	-

[a] Method B conditions: [Tris buffer] = 0.02 M; μ = 0.1 (KCl); 0.015:0.197:0.788 = THF:H_2O:DMSO(V/V); initial substrate concentration = 2.5×10^{-5} N; catalyst concentration = 2.5×10^{-4} N (of repeat units); T = 25.0°C.

[b] k_{meas} of uncatalyzed reaction.

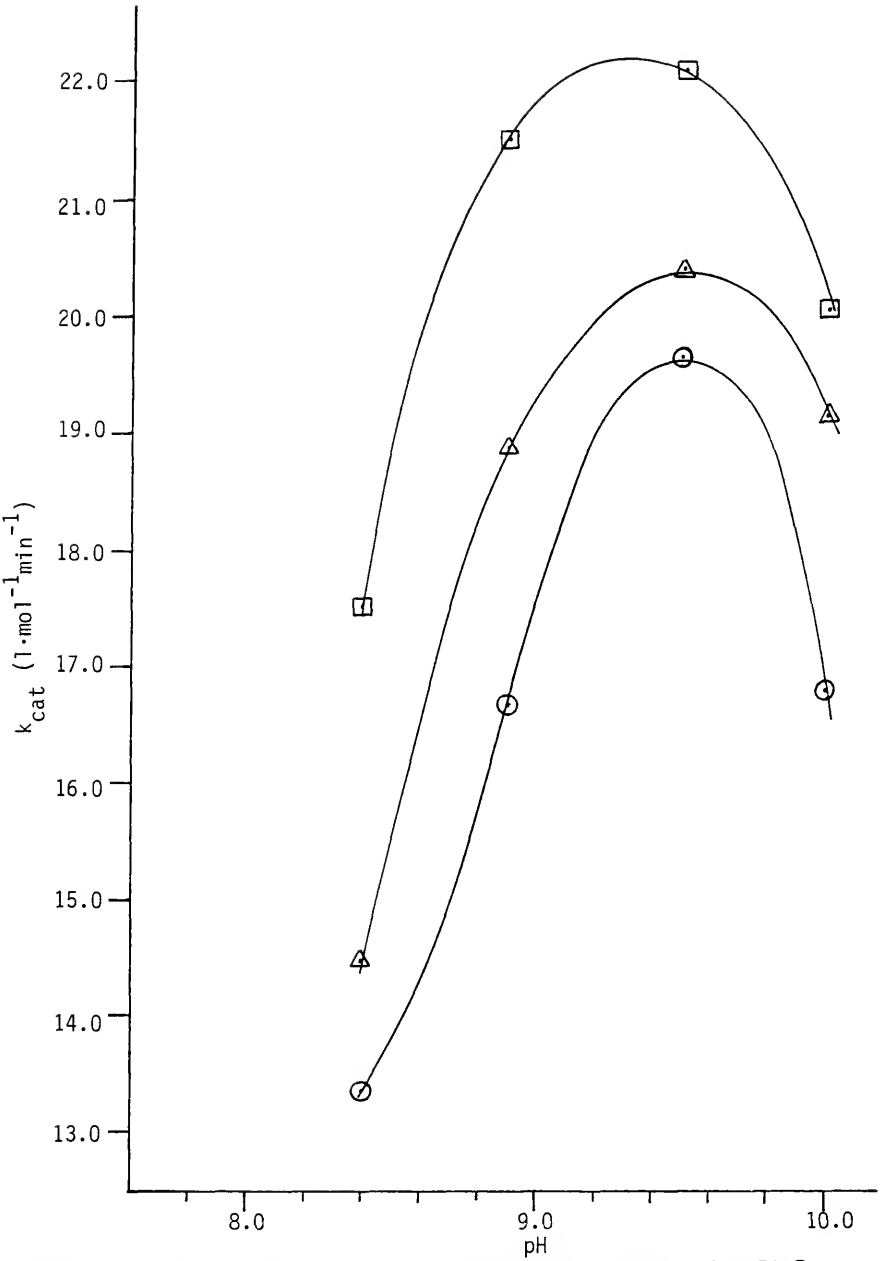


Figure 5. pH-rate profile for the esterolysis of DNPB using 59 ○, 60 △, and 62 □ as catalysts.

It can be seen that copolymer 63 exhibited little or no catalytic activity toward hydrolysis of DNPB, which implies that the phenol group or phenolate ion is catalytically inactive. All of the imidazole containing copolymers (59, 60 and 62) catalyze the esterolysis of DNPB at about the same rate. No cooperative interactions between imidazole and phenol groups were observed by comparison of the rates for 59 and 60 vs 62. Indeed, copolymer 62 was the most efficient catalyst in this group. The differences in the rate constants for 59, 60 and 62 are most likely the result of unequal concentrations of imidazole groups.

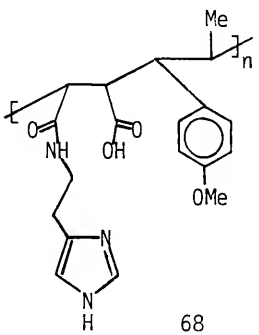
As can be seen in Figure 5, copolymers 59, 60 and 62 display a bell-shaped pH-rate profile. Such behavior is often interpreted as a result of simultaneous participation by a general acid (BH^+) and a weak base (nucleophilic or general). The concentration of BH^+ would decrease with increasing pH which would account for the decrease in k_{cat} at pH = 10.0. A reasonable candidate for BH^+ is the imidazolium ion. This argument is unsatisfactory, however, because of the extreme reactivity of DNPB, i.e., no assistance by a general acid should be required to effect hydrolysis. Also, the pKa of the 4-methylimidazolium ion is 7.61[83] which would indicate that the concentration of imidazolium ion in the polymer is very low in the pH range studied. Without a knowledge of the pKa of the imidazolium ion in the copolymers studied, this latter argument is speculative.

Another factor which might account for a bell-shaped pH-rate profile is the conformation of the copolymers in solution. If ring closure following derivatization of the maleic anhydride copolymers with histamine did not proceed quantitatively, a small number of succinamic

acid units would result giving rise to a polyelectrolyte. Alternatively, ring opening might occur under the conditions of the kinetic experiments. To check this latter hypothesis, copolymer 62 was recovered after standing in buffer solution at pH = 10.0 for 15 days. The IR spectrum of recovered 62 was identical to the spectrum of untreated copolymer, indicating that no ring opening had occurred in buffer solution.

Copolymer 62 did exhibit polyelectrolyte behavior as determined by an increase in $\eta_{sp/c}$ on dilution (Figure 6). The bell-shaped pH-rate profile might then be explained by chain expansion, allowing greater access of DNPB to imidazole residues and chain contraction at higher pH leading to a decrease in observed rate.

To test this hypothesis, compound 26 and copolymer 68 were evaluated as catalysts, Table X , Figure 7.



68

Model compound 26 exhibited a similar bell-shaped pH-rate profile, which gave evidence that this behavior could not be attributed to copolymer conformation.

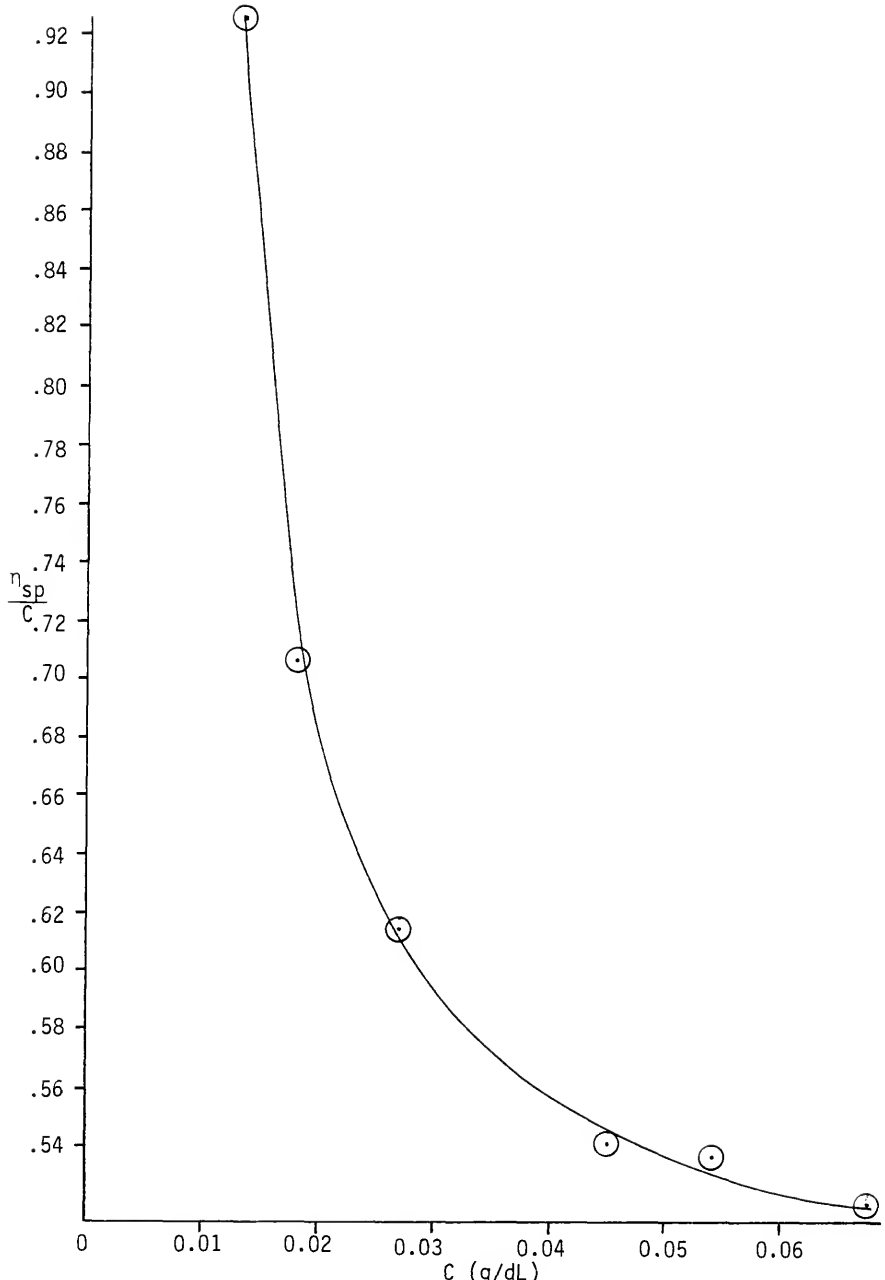


Figure 6. Plot of η_{sp}/C vs. C for copolymer 62, 0.02M Tris buffer, μ = 0.02 (KCl), pH = 9.5.

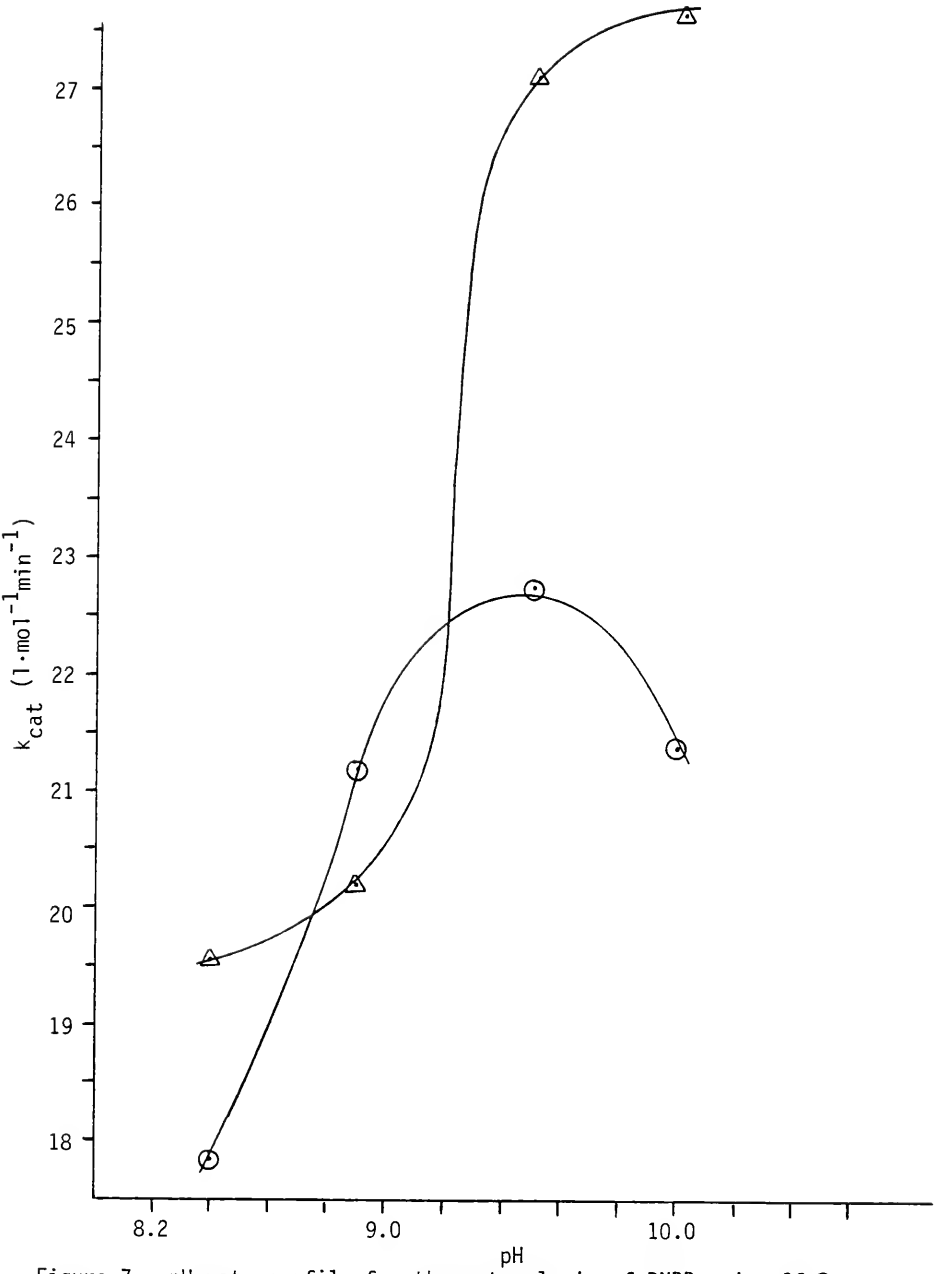


Figure 7. pH-rate profile for the esterolysis of DNPB using 26 ○ and 68 △ as catalysts.

In view of the small differences in k_{cat} with increasing pH, it became evident that the bell-shaped pH-rate profile might not be real. Indeed, there seems to be only a slight dependence of catalytic activity on pH. Therefore, the bell-shape might be the result of error associated with the determination of pH and k_{obs}.

Conclusion

Our inability to observe cooperativity between imidazole -- hydroxamic acid and imidazole -- phenol groups is surprising in light of results by other workers. Cooperativity might be expected in alternating copolymers only when a precise stereochemical fit can be achieved between substrate and functional groups. Cooperativity reported by other workers in random bifunctional copolymers might be the result of a serendipitous alignment of functional groups as a result of chain conformation.

Further work in this area should concentrate on obtaining a precise stereochemical fit between substrate and functional groups. Model compounds might provide the basis for further polymer development. Some parameters which might be modified include: the distance of functional groups from the backbone, the distance of functional groups from each other, the overall hydrophobicity of the polymer, and the nature of the functional groups themselves. One might also imitate natural enzymes by making insoluble but swellable catalysts. Soluble polymers possessing both hydrophobic and hydrophilic regions might be obtained via block copolymerization of appropriate pre-polymers. The synzyme field is full of opportunity - we have not yet scratched the surface.

APPENDIX

SELECTED ^{1}H AND ^{13}C NMR SPECTRA

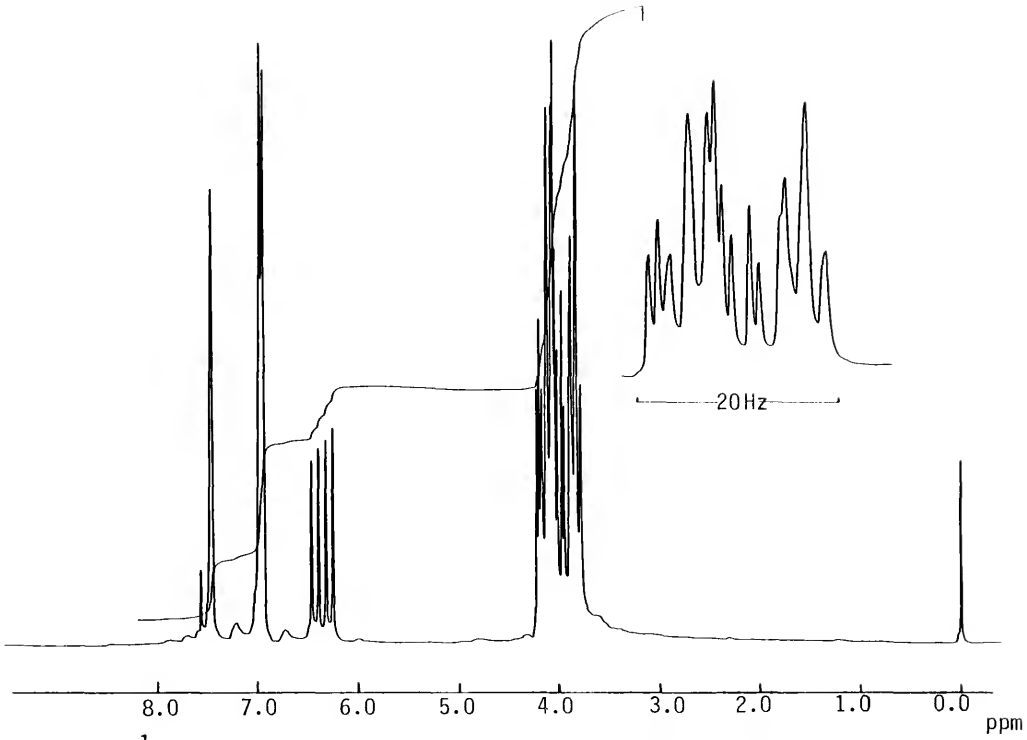


Figure 8. ^{1}H NMR spectrum of N-(β-Vinyloxyethyl)imidazole (28) in $CDCl_3$.

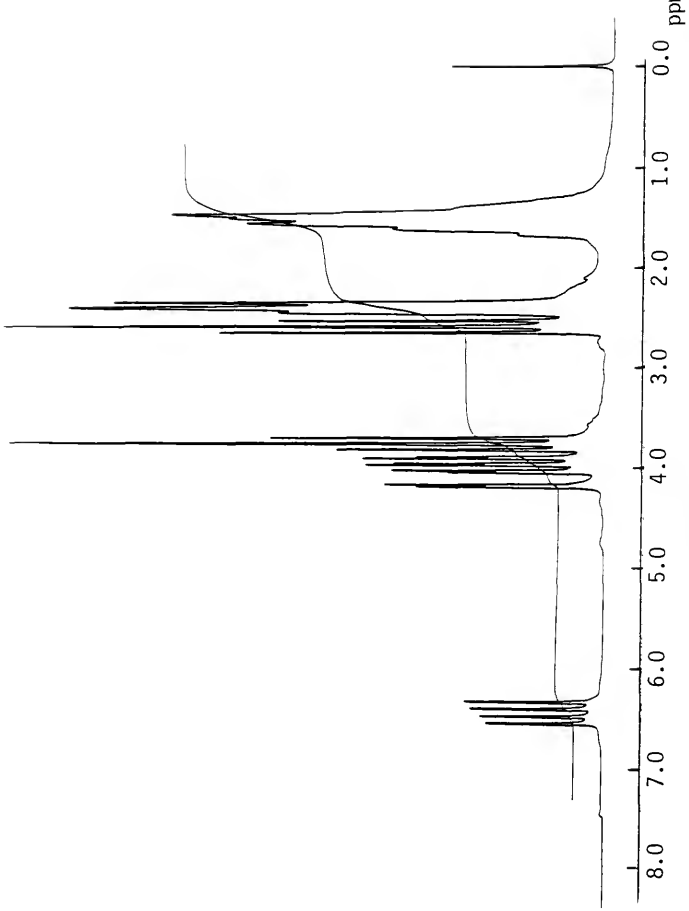


Figure 9. ^{1}H NMR spectrum of N-(β-Vinyloxyethyl)piperidine (29) in $CDCl_3$.

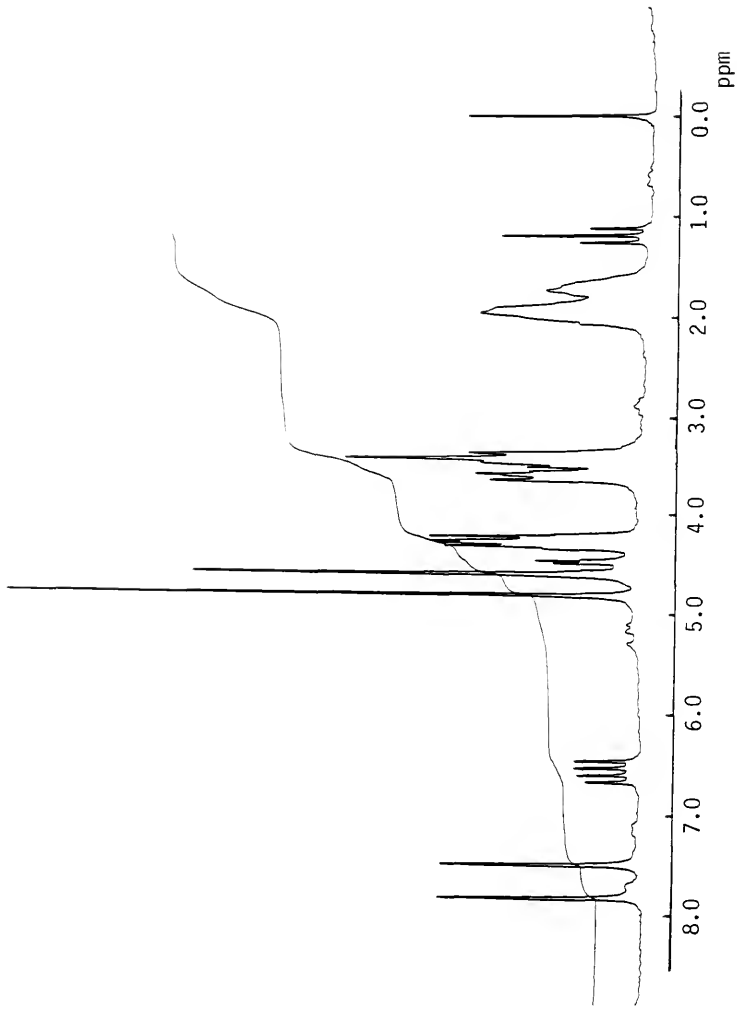


Figure 10. ^{1}H NMR spectrum of β-Vinyloxyethyl(imidazol - 4ylmethyl)piperidinium Chloride (30) in D_2O.

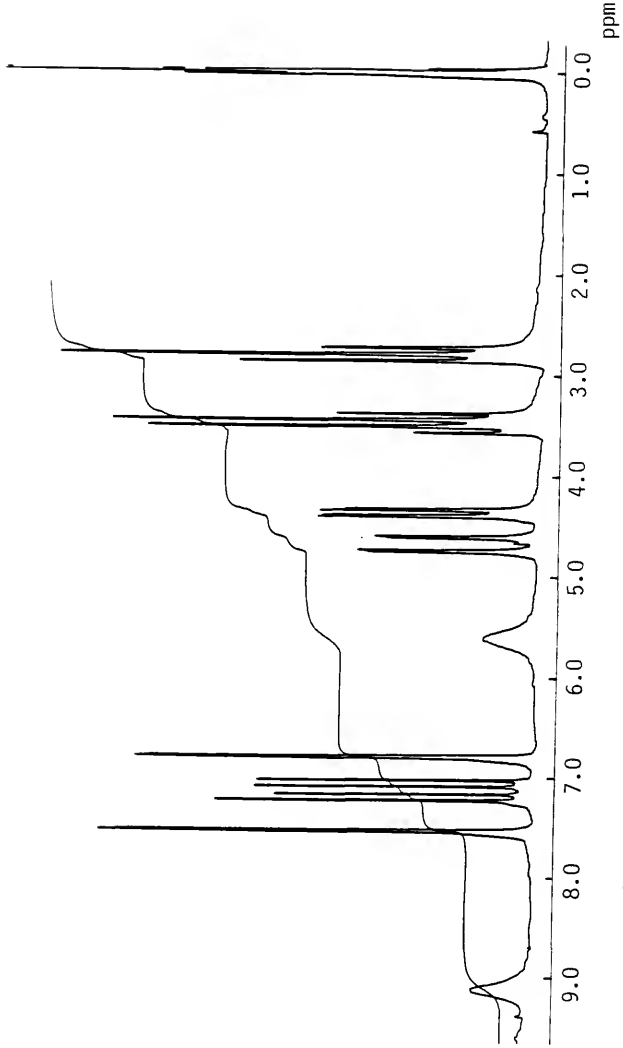


Figure 11. ^{1}H NMR spectrum of N-[(Ethenyloxy)carbonyl]-1H-imidazole-4-ethanamine (35) in $CDCl_3$.

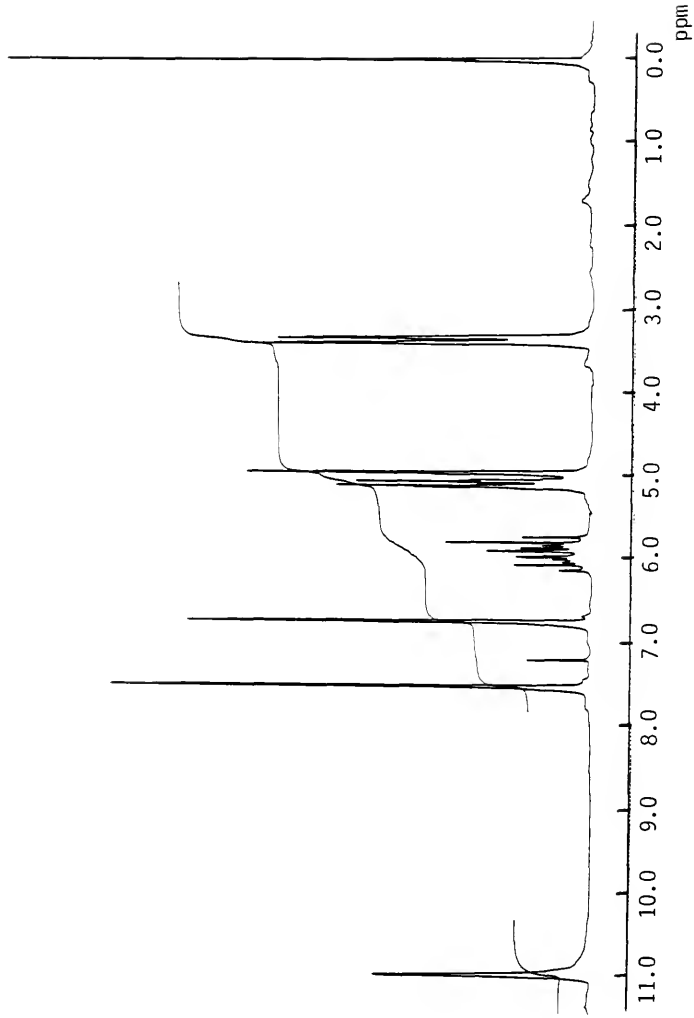


Figure 12. ^{1}H NMR spectrum of 4-Allylimidazole (38) in $CDCl_3$.

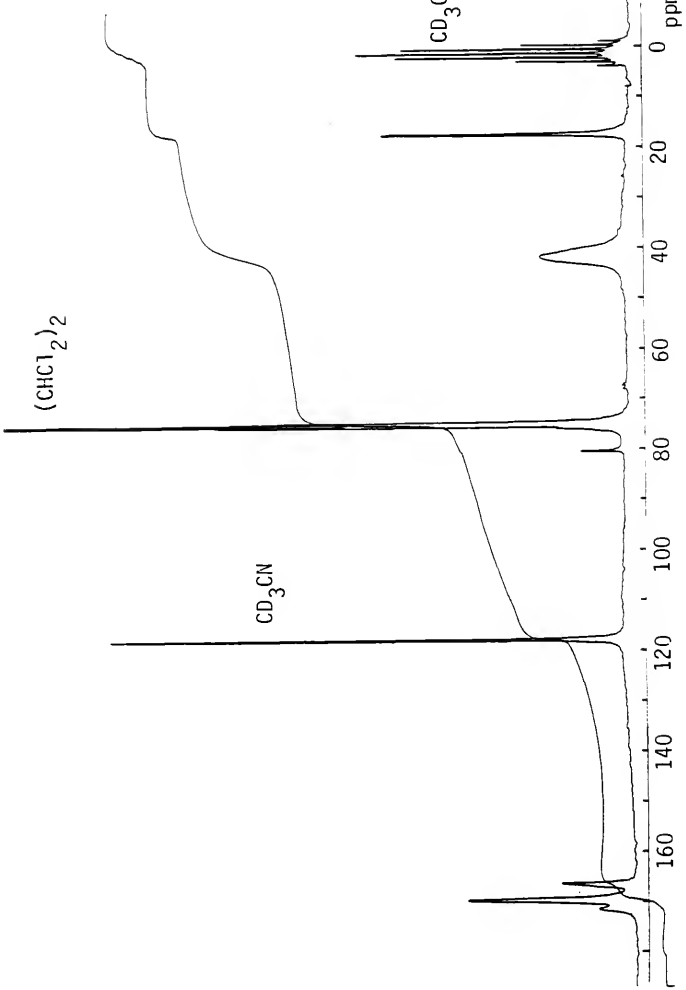


Figure 13. ^1H decoupled ^{13}C NMR spectrum of Poly(N-Acetoxymaleimide) (48) in CD_3CN-$(CHCl_2)_2$ at 60°C.

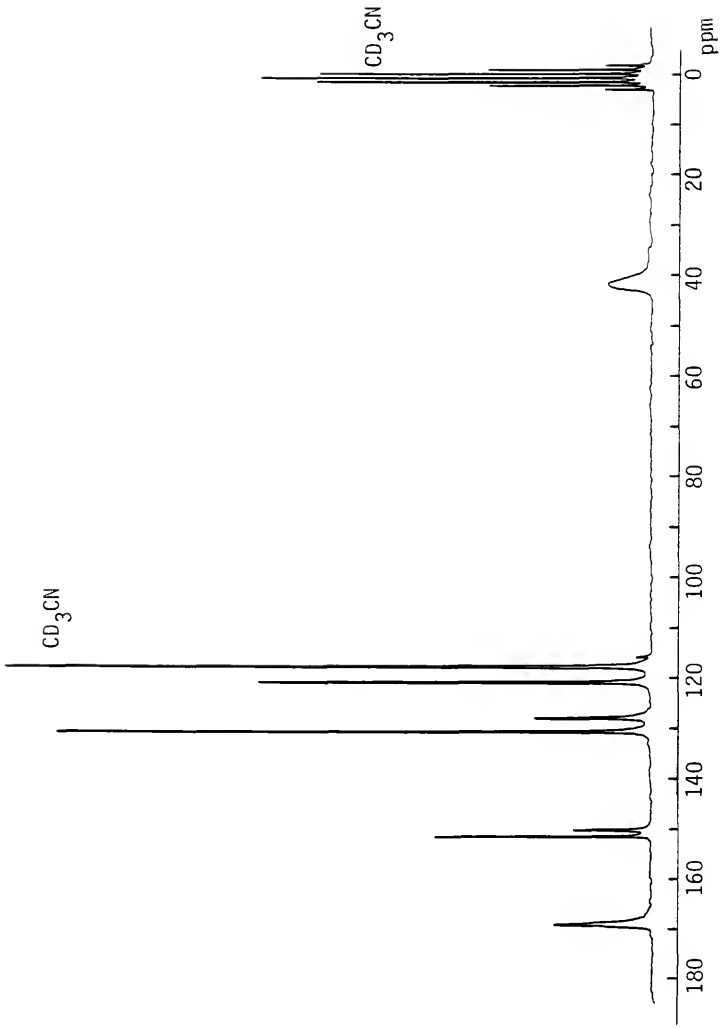


Figure 14. ^{1}H decoupled ^{13}C NMR spectrum of Poly(Phenyl N-Maleimidyl Carbonate) (49) in CD_3CN at 70°C.

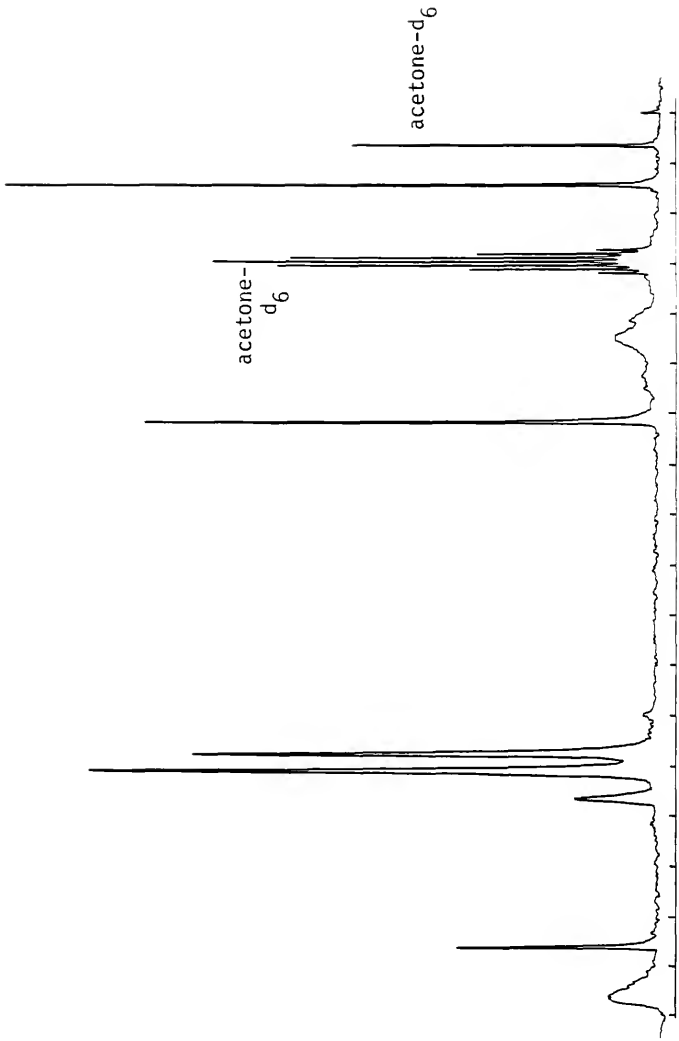


Figure 15. ^{1}H decoupled ^{13}C NMR spectrum of Poly[N-(4-Carbethoxyphenyl)maleimide] (51) in acetone-d_6 at 50°C.

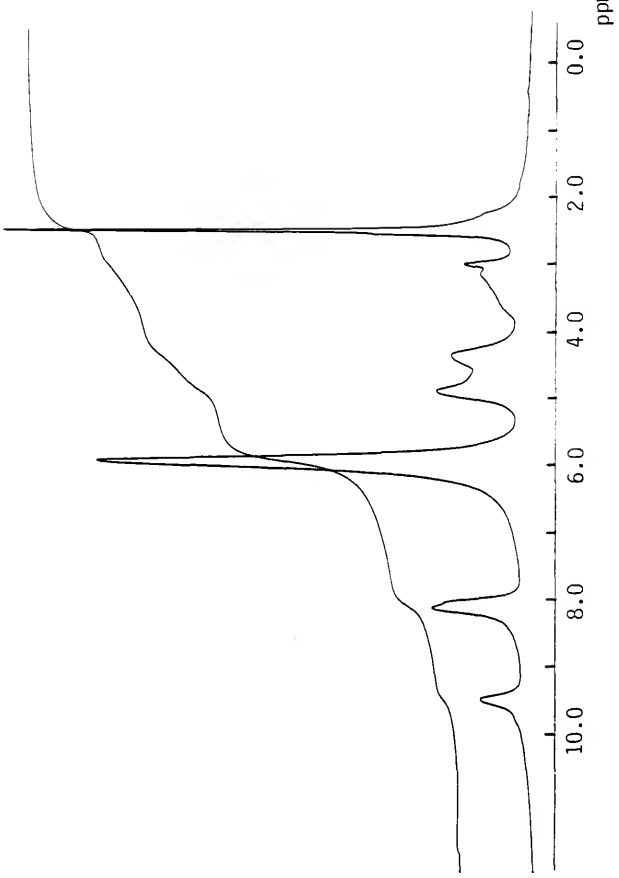


Figure 16. ^{1}H NMR spectrum of N-(β-Vinyloxyethyl)imidazole-N-Hydroxy-maleimide alternating copolymer (53) in DMSO-d_6 at 120°C.

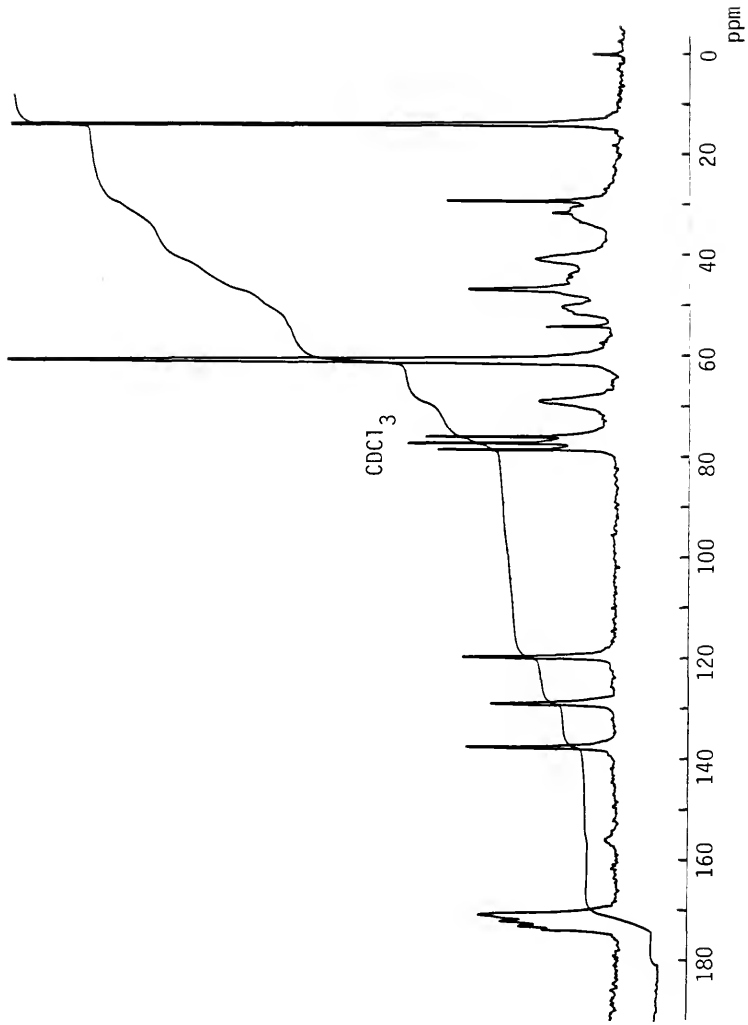


Figure 17. ^{1}H decoupled ^{13}C NMR spectrum of N-(β-Vinyloxyethyl)imidazole-Diethylfumarate copolymer (56) in $CDCl_3$.

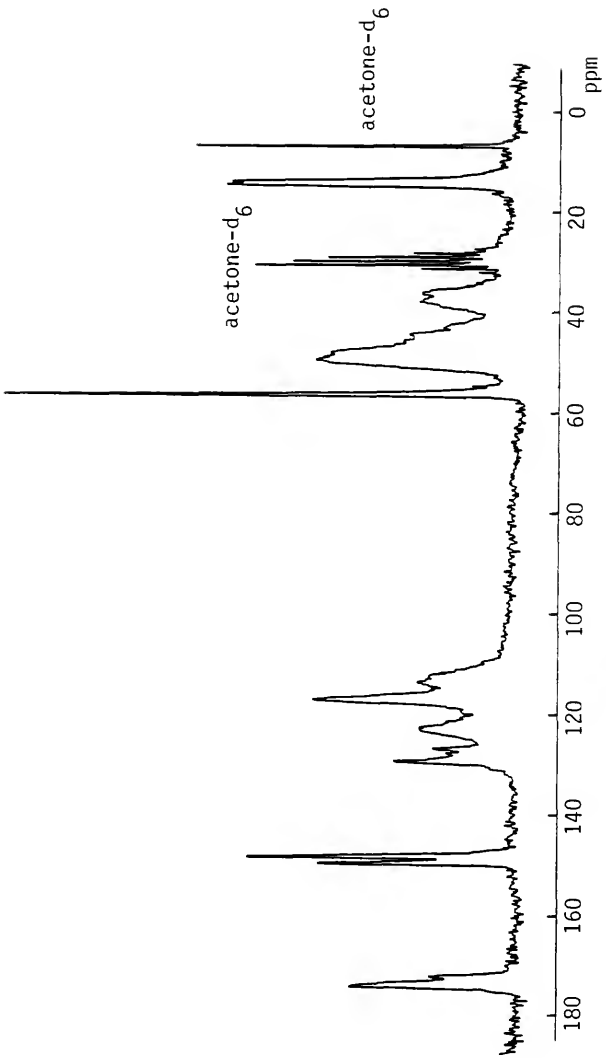


Figure 18. ^{1}H decoupled ^{13}C NMR spectrum of Isoeugenol -- Maleic Anhydride copolymer (57) in acetone-d_6 at 45°C.

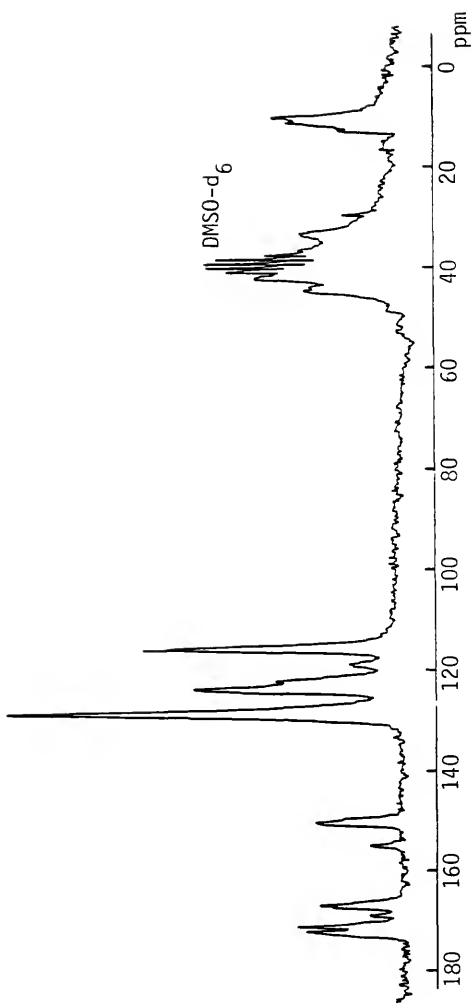


Figure 19. ^{1}H decoupled ^{13}C NMR spectrum of 2-Propenylphenol --Maleic Anhydride copolymer (58) in DMSO-d_6 at 120°c.

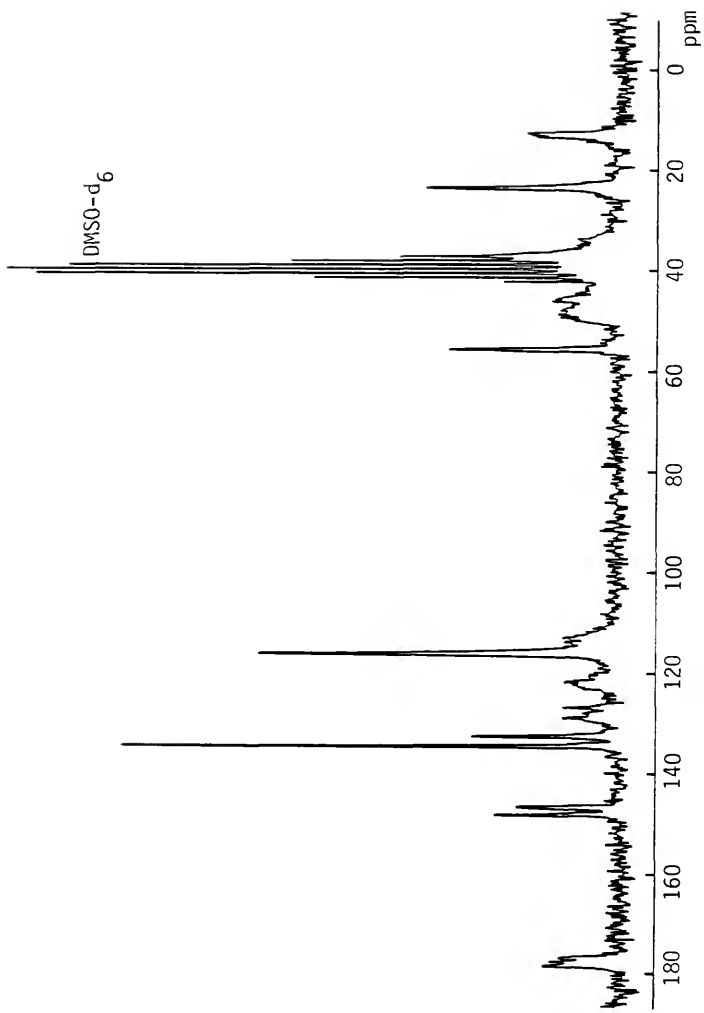


Figure 20. ^{1}H decoupled ^{13}C NMR spectrum of Isoeugenol -- N-[2-(4-Imidazolyl)ethyl] maleimide copolymer (59) in DMSO-d_6 at 110°C.

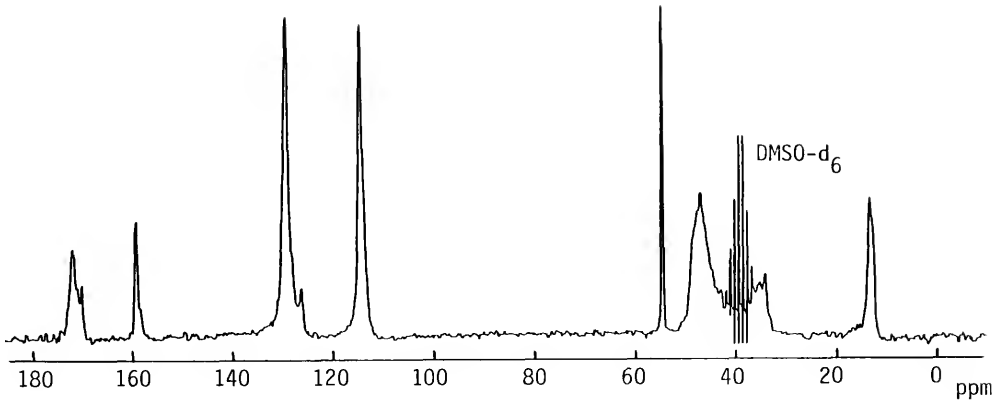


Figure 21. ^{1}H decoupled ^{13}C NMR spectrum of trans-Anethole -- Maleic Anhydride copolymer (61) in DMSO-d_6 at 110°C.

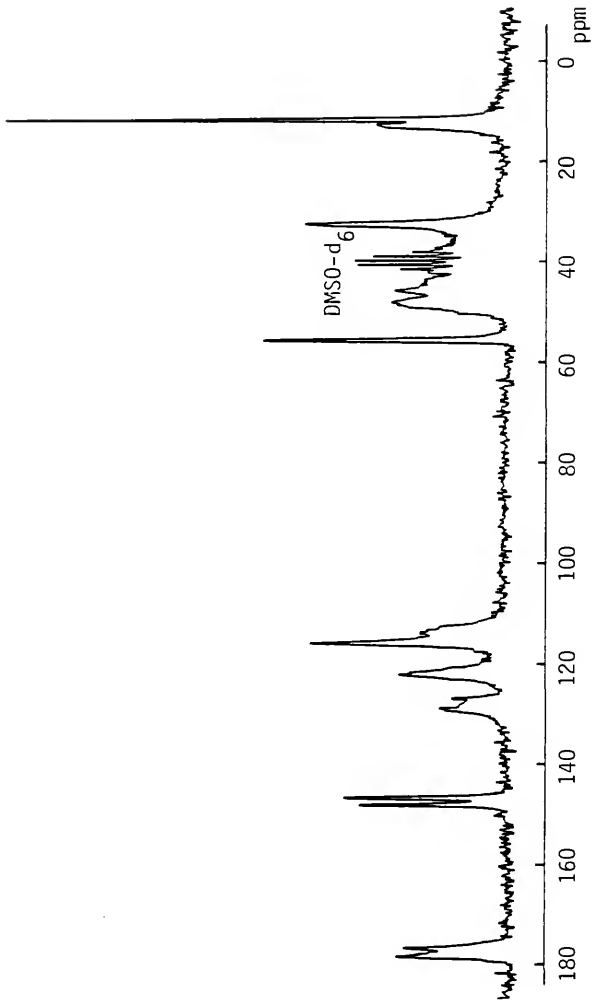


Figure 22. ^{1}H decoupled ^{13}C NMR spectrum of Isoeugenol-- N-Ethylmaleimide copolymer (63) in DMSO-d_6 at 110°C.

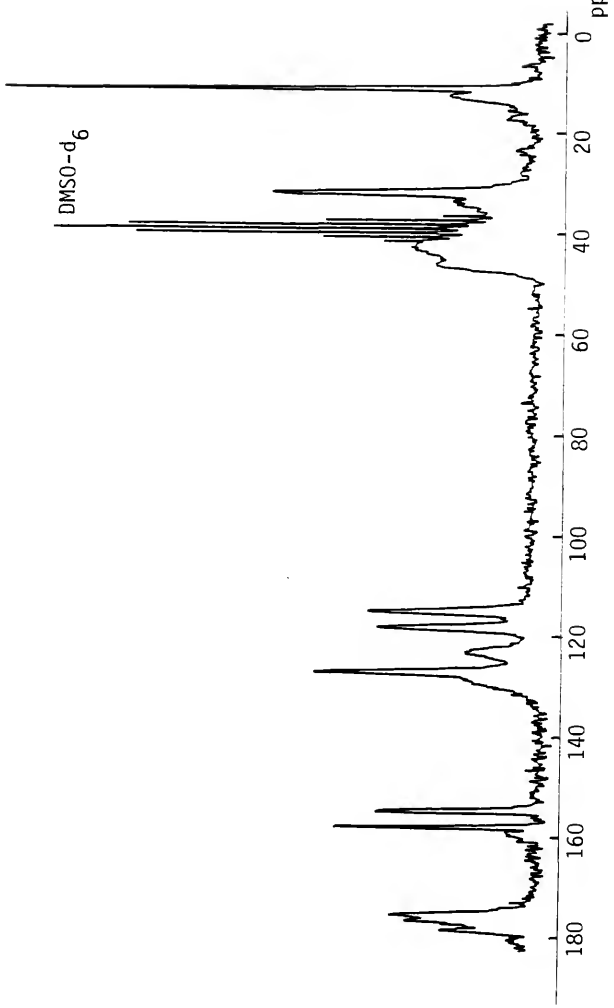


Figure 23. ^{1}H decoupled ^{13}C NMR spectrum of 2-Propenylphenol -- N-Ethylmaleimide copolymer (64) in DMSO-d_6 at 110°C.

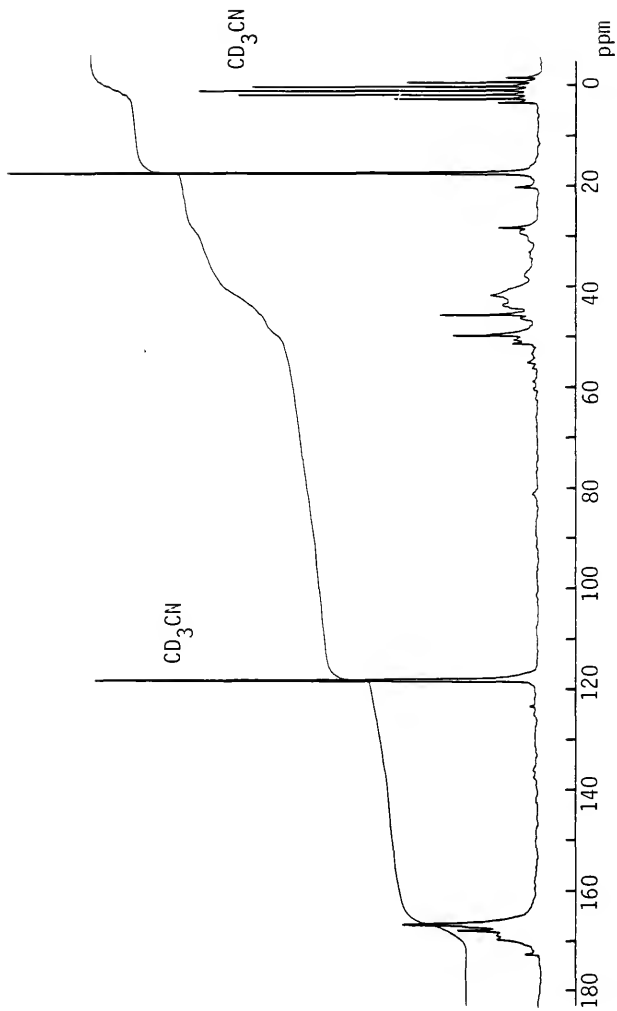


Figure 24. ^1H decoupled ^{13}C NMR spectrum of N-Acetoxymaleimide cyclotrimer (65) in CD_3CN at 70°C.

REFERENCES

1. A. Fersht, "Enzyme Structure and Mechanism," W.H. Freeman and Company, Reading and San Francisco, 1977.

2. For reviews, see:

   a. T. Kunitake and Y. Okahata in "Advances in Polymer Science," Vol. 20, H.-J. Cantow, Ed., Springer-Verlag, Berlin, 1976, p. 159.
   b. T. Shimidzu in "Advances in Polymer Science," Vol. 23, H.-J. Cantow, Ed., Springer-Verlag, Berlin, 1977, p. 55.
   c. C.G. Overberger, T.W. Smith and K.W. Dixon, J. Polym. Sci.: Symposium No. 50, 1 (1975).
   d. C.G. Overberger and S. Mitra, Pure & Appl. Chem. 51, 1391 (1979).
   e. C.G. Overberger and J.C. Salamone, Acc. Chem. Res. 2, 217 (1969).
   f. C.G. Overberger, A.C. Guteryl, Jr., Y. Kawakami, L.J. Mathias, A. Meenakshi and T. Tomono, Pure & Appl. Chem. 50, 309 (1978).

3. C.G. Overberger and H. Maki, Macromolecules 3, 214 (1970).

4. a. I.M. Klotz and V.H. Stryker, J. Am. Chem. Soc. 90, 2727 (1968).
   b. I.M. Klotz, G.P. Royer and I.S. Searper, Proc. Nat. Acad. Sci. USA 68, 263 (1971).

5. C.G. Overberger and T.W. Smith, Macromolecules 8, 407 (1975).

6. T. Kunitake and Y. Okahata, Macromolecules 9, 15 (1976).

7. a. C.G. Overberger, J.C. Salamone and S. Yaroslavsky, J. Am. Chem. Soc. 89, 6231 (1967).
   b. M.L. Bender, F.J. Kezdy and B. Zerner, J. Am. Chem. Soc. 85, 3017 (1963) and references therein.

8. J.M. Barrales-Rienda, J.I. Gonzales de la Campa and J.G. Ramos, J. Macromol. Sci.-Chem. A11, 267 (1977).

9. S. Iwatsuki and T. Itoh, Makromol. Chem. 180, 663 (1979).

10. J. Asakura, M. Yoshihara and T. Maeshima, J. Polym. Sci.: Polym. Chem. Ed. 19, 1269 (1981).

11. B.M. Culbertson, L.K. Post and A.E. Aulabaugh, Polymer Preprints 23, 1 (1982).

12. K. Olson, Ph.D. Dissertation, University of Florida, 1981.

13. Merck & Co., Inc., "Deuterated NMR Solvents - Handy Reference Data," Merck & Co., Inc., Quebec, 1978.

14. A.J. Gordon and R.A. Ford, "The Chemists Companion: A Handbook of Practical Data, Techniques and References," John Wiley and Sons, New York, 1972.

15. M. Narita, T. Teramoto and M. Okawara, Bull. Chem. Soc. Jp. 44, 1084 (1971).

16. "Aldrich Catalog Handbook of Fine Chemicals - 1981-1982," Aldrich Chemical Co., Milwaukee, 1980, p. 427.

17. M. Akiyama, K. Shimizu, S. Aiba and F. Banba, J. Chem. Soc. Perkin Trans. I, 2122 (1980).

18. Sadtler NMR Spectrum #14580.

19. V.S. Ivanov, V.K. Smirnova and T. Yung, Metody Poluch. Khim. Realctivov Prep., 113 (1967); Chem. Abstr. 71, 12489v, (1969).

20. O. Keller and J. Rudinger, Helv. Chim. Acta 58, 531 (1975).

21. Dr. Huey Pledger, Jr., Visiting Research Scientist, University of Florida.

22. L.A. Holt, S.J. Leach and B. Milligan, Aust. J. Chem. 21, 2115 (1968).

23. P. Fournari, P. de Cointet and E. Laviron, Bull. Soc. Chim. Fr., 2438 (1968).

24. R.L. Goette, M.S. Thesis, University of Florida, 1950.

25. U. Tonellato, J. Chem. Soc. Perkin Trans. II, 771 (1976).

26. H. Tabor and E. Mosettig, J. Biol. Chem. 180, 703 (1949).

27. M. Windholz, Ed., "The Merck Index," Vol. 9, Merck & Co., Inc., Rahway, NJ, 1976.

28. J.R. Totter and W.J. Darby in "Organic Syntheses, Collective Volume 3," J. Wiley & Sons, Inc., New York, 1955, p. 460.

29. R.A. Turner, C.F. Huebner and C.R. Scholz, J. Am. Chem. Soc. 71, 2801 (1949).

30. J. Pyman, J. Chem. Soc. 99, 675 (1911).

31. W.J. Eilbeck, F. Holmes and A.E. Underhill, J. Chem. Soc. A, 757 (1967).

32. J.A. Welleman, F.B. Hulsbergen, J. Verbiest and J. Reedijk, J. Inorg. Nucl. Chem. 40, 143 (1978).

33. C.G. Begg, M.R. Grimmett and P.D. Wethey, Aust. J. Chem. 26, 2435 (1973).

34. J.E. Stambaugh and R.W. Manthei, J. Chromatog. 31, 128 (1967).

35. D.S. Tarbell in "Organic Reactions Vol. II," R. Adams, W.E. Bachmann, L.F. Fieser, J.R. Johnson and H.R. Snyder, Eds., J. Wiley & Sons, Inc., New York, 1944, p. 1.

36. Y. Shirota, M. Yoshimura, A. Matsumoto and H. Mikawa, Macromolecules 7, 4 (1974).

37. "Aldrich Catalog Handbook of Fine Chemicals - 1981-1982," Aldrich Chemical Co., Milwaukee, 1980, p. 491.

38. R.C. Weast, Ed., "CRC Handbook of Chemistry and Physics, 60th Ed.," CRC Press, Inc., Boca Raton, FL, 1979, p. C-87.

39. A. Mobley, University of Florida.

40. C.G. Overberger, R.C. Glowaky, T.J. Pacansky and K.N. Sannes in "Macromolecular Syntheses, Collective Vol. I," J.A. Moore, Ed., John Wiley and Sons, Inc., New York, 1978, p. 619.

41. J.H. Espenson, "Chemical Kinetics and Reaction Mechanisms," McGraw-Hill, Inc., New York, 1981, p. 24.

42. T. Kunitake and Y. Okahata, Bioorg. Chem. 4, 136 (1975).

43. T. Kunitake, Y. Okahata and R. Ando, Bull. Chem. Soc. Jp. 47, 1509 (1974).

44. T. Kunitake and Y. Okahata, Chem. Lett., 1057 (1974).

45. M.K. Hargreaves, J.G. Pritchard and H.R. Dave, Chem. Reviews 70, 439 (1970).

46. G. Hunter and J.A. Nelson, Can. J. Resh. 19B, 296 (1941).

47. C.A. Buehler and D.E. Pearson, "Survey of Organic Synthesis," Wiley Interscience, New York, 1970, p. 416.

48. L.F. Fieser and M. Fieser, "Reagents for Organic Synthesis," John Wiley and Sons, Inc., New York, 1967, p. 1030.

49. N.E. Searle, U.S. Patent, 2,444,536, July 6, 1948; Chem. Abstr. 42, 7340 (1948).

50. M. Narita, M. Akiyama and M. Okawara, Bull. Chem. Soc. Jp. 44, 437 (1971).

51. S.R. Sandler and W. Karo, "Organic Functional Group Preparations, Vol. III," Academic Press, New York, 1971, p. 406.

52. D.E. Ames and T.F. Grey, J. Chem. Soc., 631 (1955).

53. V.S. Ivanov, V.K. Smirnova, A.E. Semenova and T. Yure, J. Org. Chem., USSR 1, 1729 (1965); Chem. Abstr. 64, 586g (1966).

54. M. Akiyama, K. Shimizu and M. Narita, Tet. Lett., 1015 (1976).

55. C.G. Overberger and H. Maki, Macromolecules 3, 214 (1970).

56. C.G. Overberger and H. Maki, Macromolecules 3, 220 (1970).

57. D.M. Blow and T.A. Steitz, Ann. Rev. Biochem. 39, 716 (1970).

58. A. Winston and D. Kirchner, Macromolecules 11, 597 (1978).

59. R.J. Cotter, C.K. Sauers and J.M. Whelan, J. Org. Chem. 26, 10 (1961).

60. L.E. Coleman, J.F. Bork and H. Dunn, Jr., J. Org. Chem. 24, 135 (1959).

61. F.E. King. J.W. Clark-Lewis, R. Wade and W.A. Swindin, J. Chem. Soc., 873 (1957).

62. G.B. Butler and A. Zampini, J. Macromol. Sci.-Chem. A11, 491 (1977).

63. W.H. Watanabe and L.E. Conlon, J. Am. Chem. Soc. 79, 2828 (1957).

64. T. Alfrey, Jr. and C.C. Price, J. Polym. Sci. 2, 101 (1947).

65. R.Z. Greenley, J. Macromol. Sci.-Chem. A14, 427 (1980).

66. J.W. Black and C.R. Ganellin, Experientia 30, 111 (1974).

67. F.B. Stocker, M.W. Fordice, J.K. Larson and J.H. Thorstenson, J. Org. Chem. 31, 2380 (1966).

68. T. Wagner-Jauregg and Q. Ahmed, Helv. Chim. Acta 56, 1406 (1973).

69. T. Wagner-Jauregg and Q. Ahmed, Helv. Chim. Acta 57, 1871 (1974).

70. M. Akiyama, M. Narita and M. Okawara, J. Polym. Sci., Part A-1 7, 1299 (1969).

71. D.M. Grant and E.G. Paul, J. Am. Chem. Soc. 86, 2984 (1964).

72. J.C. Salamone, W. Volksen, A.P. Olson and S.C. Israel, Polymer 19, 1157 (1978).

73. F.W. Billmeyer, Jr., "Textbook of Polymer Science, 2nd Ed.," John Wiley and Sons, Inc., New York, 1971, p. 363.

74. F.A. Cotton and R. Francis, J. Am. Chem. Soc. 82, 2986 (1960).

75. Y. Shirota, M. Yoshimura, A. Matsumoto and H. Mikawa, Macromolecules 7, 4 (1974).

76. S.R. Sandler and W. Karo, "Polymer Syntheses II," Academic Press, New York, 1977, p. 214.

77. N.D. Field and D.H. Lorenz in "Vinyl and Diene Monomers-Part 1," E.C. Leonard, Ed., Wiley-Interscience, New York, 1970, p. 396.

78. C.E. Schildknecht in "Kirk-Othmer Encyclopedia of Chemical Technology-2nd Ed.," Vol. 21, Wiley-Interscience, New York, 1970, p. 412.

79. C.E. Schildknecht, C.H. Lee and W.E. Maust in "Macromolecular Syntheses, Collective Volume I," J.A. Moore, Ed., John Wiley and Sons, Inc., New York, 1978, p. 113.

80. G. Hardy, K. Nyitrai and F. Cser in "Macromolecular Syntheses, Collective Volume I," J.A. Moore, Ed., John Wiley and Sons, Inc., New York, 1978, p. 669.

81. S.L.N. Seung and R.N. Young, J. Polym. Sci.-Polym. Lett. Ed. 16, 367 (1978).

82. S. Iwabuchi, K. Kojima, T. Nakahira and H. Hosoya, Makromol. Chem. 177, 1643 (1976).

83. F. Schneider, Z. Physiol. Chem. 338, 131 (1964); Chem. Abstr. 62, 11905 (1965).

BIOGRAPHICAL SKETCH

David Paul Vanderbilt was born on January 18, 1954, in Easton, PA. He was a 1971 graduate of Wilson Boro High School, Easton, PA. The author received the B.S. degree in chemistry from the Pennsylvania State University, University Park, PA,in 1975.

Mr. Vanderbilt enrolled in the Graduate School at the University of Florida in September, 1976. He received the M.S. degree in chemistry in August, 1979, under the auspices of Professor Merle A. Battiste. While pursuing M.S. and Ph.D. degrees at U.F., the author served as a teaching and research assistant in the Department of Chemistry. He is a member of the American Chemical Society.

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

George B. Butler, Chairman
Professor of Chemistry

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Merle A. Battiste
Professor of Chemistry

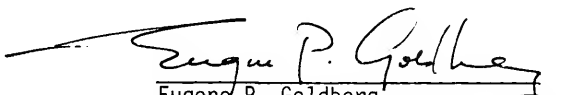
I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Thieo E. Hogen-Esch
Professor of Chemistry

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

Gus J. Palenik
Professor of Chemistry

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.



Eugene P. Goldberg
Professor of Materials Science
and Engineering

This dissertation was submitted to the Graduate Faculty of the Department of Chemistry in the College of Liberal Arts and Sciences and to the Graduate Council, and was accepted as partial fulfillment of the requirements for the degree of Doctor of Philosophy.

December, 1982

Dean for Graduate Studies
and Research